MONOGRAPHS

IN

Medicine

MONOGRAPHS IN MEDICINE

SERIES 1

EDITOR

WILLIAM B. BEAN, M.D.
Professor and Head of the Department of Medicine, State University of Iowa

ASSOCIATE EDITORS

MORTON HAMBURGER, M.D.
Associate Professor of Medicine, University of Cincinnati School of Medicine

JOHN A. LEUTSCHER, JR., M.D.
Associate Professor of Medicine, Stanford University School of Medicine

STEWART WOLF, M.D.
Professor and Head of the Department of Medicine, University of Oklahoma School of Medicine



BALTIMORE
THE WILLIAMS & WILKINS COMPANY
1952

Copyright, 1952
The Williams & Wilkins Company

Made in the United States of America

Printed for
The Williams & Wilkins Company
by the
Waverly Press, Inc
Baltimore 2, Md., U.S.A

Preface

The launching of a new series of collected monographs demands some word of justification and purpose. Knowledge of advance in medicine may be gained in several ways. The first is travel, which permits the direct exchange of ideas, discussion of work in progress, and gives the best yield when there is ample time for detailed cross questioning. But travel is impractical as a sustained program, and impossible for a large number of physicians. Another way is attendance at medical meetings. It is especially stimulating where reports of investigation and observation are presented. It enables one to judge the author by personal contact.

The third way is to read the weekly, monthly or quarterly journals which proliferate in such profusion; and tax the wit and patience of the most steadfast reader Some contributors to our medical journals fail to relate their work to what has gone before, plunging in without clear beginning or continuity (and we all must stand on the shoulders of those who have preceded us). Add to this the slovenly tendency to leave many loose ends, the failure to interpret data, or failure to seek significance except perhaps statistical significance, and we have before us the raw material of chaos A process of culling yields a variable reward, but there is necessarily much waste of time and energy. Almost inevitably, by delays in preparation, editing, printing and circulating the material in most journals is a year behind its presentation at a meeting, which may in turn be a year behind actual work

The monograph best serves its purpose when it combines the most useful features of these three methods of obtaining knowledge, and, as proposed for this and later volumes, may prove a useful compromise between the journal focused on the temporary present, and the fully comprehensive volume that purports to cover the entire field of knowledge. The aim is to provide a series of articles varying widely but in fields germane to general medicine. The treatment, though critical, may not necessarily be final It will be digested enough so that its utility is clear and its significance understood

Medical progress is not delayed by taking time out to survey the scene from the medium hills, neither staying on the flats of day-by-day affairs nor climbing the high mountain for the grand and inclusive spectacle The purpose, though modest, is a necessary one if we are to retrieve order in medical thought among the lush jungles of so many fast-flowering subspecialties For good monographs there is always ready need.

My task has been lightened substantially and made pleasant by the

active work of the associate editors, Dr. Morton Hamburger, Dr. John Luetscher and Dr Stewart Wolf. With them and the contributors I share responsibility as well as eager hope for these and later *Medical Monographs*

WILLIAM BENNETT BEAN

Iowa City, Spring 1952

Contributors

HENRY ARANOW, JR., M.D., Med.Sc.D.
Associate in Medicine, Columbia University College of Physicians and Surgeons; Presbyterian Hospital, New York City (*page 179*).

WILLIAM BENNETT BEAN, M.D.
Professor and Head of the Department of Medicine, State University of Iowa, Iowa City (*page 22*).

CHARLES G. CHILD, III, M.D.
Attending Surgeon, New York Hospital; Associate Professor of Clinical Surgery, Cornell University Medical College (*page 132*)

FLOYD W. DENNY, M.D.
Instructor in Pediatrics, University of Minnesota School of Medicine (*page 295*).

CHARLES T. DOTTER, M.D.
Assistant Professor of Radiology, Cornell University Medical College, Assistant Attending Radiologist, New York Hospital (*page 103*)

HENRY E. HAMILTON, M.D.
Assistant Professor of Internal Medicine, College of Medicine, University Hospitals, State University of Iowa (*page 315*)

DAVID A. KARNOFSKY, M.D.
Chemotherapy Service, Memorial Center for Cancer and Allied Diseases, New York (*page 582*)

BYRD S. LEAVELL, M.D.
Associate Professor of Medicine, University of Virginia School of Medicine (*page 451*)

WILLIAM A. MACILWAINE, M.D.
Research Fellow in Medicine of the American College of Physicians, University of Virginia School of Medicine (*page 451*)

MAX MICHAEL, JR., M.D.
Chief of Medical Service, Lawson V.A. Hospital, Associate Professor of Clinical Medicine, Emory University School of Medicine (*page 271*).

JOHN F. MUELLER, M.D.
Assistant Professor of Medicine, University of Cincinnati School of Medicine (*page 519*).

MARY ANN PAYNE, PH D., M D.
Assistant Professor of Clinical Medicine, Cornell University Medical College; Assistant Attending Physician, New York Hospital (*page 132*).

FRED PLUM, M.D.
Instructor in Medicine, Cornell University Medical College; Executive Officer and Assistant to the Director, Second (Cornell) Division, Neurological Service, Bellevue Hospital, N. Y.; Assistant Visiting Physician, Bellevue Hospital (*page 225*).

CHARLES H. RAMMELKAMP, JR., M.D.
Professor of Medicine, Associate Professor of Preventive Medicine, Western Reserve University School of Medicine (*page 295*).

R WALTER SCHLESINGER, M.D.
Associate Member, Division of Infectious Diseases, The Public Health Research Institute of the City of New York, Inc (*page 380*).

L. H SCHMIDT, PH D
Director, Christ Hospital Institute of Medical Research, Cincinnati (*page 348*)

ISRAEL STEINBERG, M D
Assistant Clinical Professor of Radiology and Medicine, Cornell University Medical College; Assistant Attending Radiologist and Assistant Attending Physician, The New York Hospital, Attending Physician and Chief of Chest Clinic, Sydenham Hospital, Attending Consultant, Chest Diseases, V.A Hospital, Bronx, Consultant in Angiocardiography, U.S N.H , St Albans, New York (*page 103*)

RICHARD W VILTER, M.D
Associate Professor of Medicine, University of Cincinnati School of Medicine (*page 519*)

WILLIAM W WALLACE, M D.
Assistant Professor of Pediatrics, Harvard Medical School; Physician, Children's Medical Center, Boston (*page 66*)

STEWART WOLF, M.D
Professor and Head of the Department of Medicine, University of Oklahoma School of Medicine, Oklahoma City (*page 1*).

Contents

Talking with the Patient*

STEWART WOLF, M.D.

GENERAL CONSIDERATIONS AND OBJECTIVES

The basis of any medical practice is communication with the patient and yet in the curriculum of most medical schools the student learns the art of communication only incidentally. Skill in communication is as fundamental a requirement for the physician as is knowledge of anatomy or chemistry. Furthermore, the growing awareness of the relevance of life stress to bodily disease has emphasized the need for the general physician and the medical specialist to understand their patients as people and to evaluate their reactions to other people and events in their lives. The question "How to do this?" has prompted the writing of the present treatise. Methods involved in talking with the patient, eliciting pertinent data, catching the significance of things said or left unsaid, and turning discussion into treatment will be illustrated by direct quotation from actual interviews with patients, recorded and transcribed verbatim.

APPROACH TO THE PATIENT

Success or failure in communication often depends heavily upon the patient's initial impression of the physician Repeatedly one hears patients say of a physician, "He's such a busy man I didn't want to burden him with my troubles" or, "He didn't have time to talk to me " The physician who, even unintentionally, has given his patient such an impression has placed himself at a disadvantage and has perhaps denied himself important diagnostic data. It is the first rule of good communication that the physician appear unhurried

Other statements frequently made by patients who have left a physician are, "I was a little afraid of him" or, "He didn't seem interested" or, "He seemed to be having troubles of his own " These remarks imply, of course, a defect in the attitude of the physician who was unable to put the patient at his ease and inspire his confidence These are important matters since good communication requires the patient's cooperation Skill in gaining the patient's confidence is no mere technical matter It is achieved on the basis

* Supported by the Commonwealth Fund, The National Heart Institute of the National Institutes of Health, Public Health Service, and Medical Research and Development Board, Department of the Army

Abstracted and adapted from a forthcoming book, "Talking with the Patient" by Stewart Wolf, to be published by Paul B Hoeber, Medical Book Division of Harper Bros , New York

of the physician's personal qualifications. First, he must have an interest in and sympathy with people and their problems. More than that he must believe in their potential for constructive adjustment. He needs to be able to "give out" encouragement and support even in the face of the patient's hostility. It is an experience of everyday practice and part of the therapeutic process that the patient expresses vicariously toward the physician in various direct and indirect ways the hostility he feels toward parents, wife or boss. The physician needs patience. He must be willing to carry responsibility while his patient is slowly working through misinterpretations and poor decisions to a better adjustment. He must often worry along with little progress or settle for a limited objective realizing that rejecting the patient or "giving him up as a bad job" would be destructive The physician must be willing to listen and to spend the time and energy which that requires.

Finally, the physician needs restraint. He must be particularly careful not to express contempt, ridicule or disapproval directly or indirectly by word or gesture. He must avoid pressing a course of action which the patient may not be able or ready to accept, however obvious it may seem to the physician. Especially, he must assiduously avoid using his patient to work out his own problems and conflicts or dominating him to enhance his own feeling of security (1).

Few of us can meet in full these stiff personal requirements but, nevertheless, each one is important. Together they spell maturity of character, emotional balance and self discipline They are qualities sought for in candidates for admission to medical schools, and are not necessarily reflected in college grades or in tests of "aptitude" (2) In the curriculum of the medical schools it will become increasingly important to cultivate in the students their natural humanitarian tendencies and their interest in the general welfare of their fellow man. The physician's role in his community has broadened. More and more he is concerned with the personal problems of his patients, their attitudes and aspirations Talking with his patient the physician finds that these matters are highly pertinent to health and disease.

The precise manner in which the physician greets his patient is a matter of personal preference However, many physicians in their desire not to be austere or forbidding may assume a casual or jaunty air with cigarette in hand and feet up. Such exaggerated and unbecoming informality is usually ineffective, if indeed not actually disconcerting to a patient who hopes his problems will be taken seriously

HISTORY TAKING

In the physician's first contact with the patient he usually "takes a history" and thus sets the stage for all subsequent talking with the patient.

Although experienced clinicians consider the history to be their most important aid in diagnosis, it would appear from a recent study of the performance of fourth year students in "history taking" that the average student goes out into his medical practice with only a hazy knowledge of communication. These students were inclined to consider the history as a technique capable of being standardized rather than as an instrument requiring manipulation and effective only in proportion to the skill of the operator. There was a widespread impression among the students that if one was careful to ask the prescribed number of questions one should come out with the answer. Few recognized the history as an inquiry in which one thing leads to another. Few were aware that there is no such thing as a complete history but that each history differs from every other depending on the nature of the patient and his illness. It is important to realize, however, that the development of pertinent data in a history does not preclude thoroughness. The deficiencies displayed by the students studied were surprisingly uniform (3). They may be grouped as follows:

Errors of Commission

a. As already indicated, most students took the history in a uniform fashion asking questions learned from an outline By seeking rapid accurate answers to such a prepared list of questions and failing to frame their questions in view of replies to earlier ones, they lost the chance to make of the history an exploration whose direction is suggested by each previous step. Moreover, the students failed to take into account the possibility that many questions may elicit answers which may be politely or protectively misleading, or entirely incorrect

b By forcing the patient to express his "chief complaint" in a few fully quotable words they often failed to get on the right track. Sometimes the chief complaint as recorded by the student contained a plausible excuse for the hospital visit, but did not reflect the patient's real reasons for seeking help Since the handling of subsequent portions of the history depends inevitably on the content of the "chief complaint", this inadequacy often led to a misdirected and largely irrelevant inquiry.

c The students did too much of the talking themselves, phrasing questions in such a manner that they could be answered "yes or "no" This not only deprived the observer of the shades of meaning which a patient could communicate by a less direct answer but denied him the important leads frequently inherent in a patient's relatively free description of his difficulties

d Often the opposite error was made at another time in the interview by the same student who allowed his patient to engage in a long circumstantial account of details of his symptoms and thus failed to bring the general problem into focus Occasionally the interview assumed the tone of

of the physician's personal qualifications. First, he must have an interest in and sympathy with people and their problems More than that he must believe in their potential for constructive adjustment. He needs to be able to "give out" encouragement and support even in the face of the patient's hostility. It is an experience of everyday practice and part of the therapeutic process that the patient expresses vicariously toward the physician in various direct and indirect ways the hostility he feels toward parents, wife or boss. The physician needs patience He must be willing to carry responsibility while his patient is slowly working through misinterpretations and poor decisions to a better adjustment. He must often worry along with little progress or settle for a limited objective realizing that rejecting the patient or "giving him up as a bad job" would be destructive. The physician must be willing to listen and to spend the time and energy which that requires.

Finally, the physician needs restraint. He must be particularly careful not to express contempt, ridicule or disapproval directly or indirectly by word or gesture. He must avoid pressing a course of action which the patient may not be able or ready to accept, however obvious it may seem to the physician. Especially, he must assiduously avoid using his patient to work out his own problems and conflicts or dominating him to enhance his own feeling of security (1).

Few of us can meet in full these stiff personal requirements but, nevertheless, each one is important Together they spell maturity of character, emotional balance and self discipline. They are qualities sought for in candidates for admission to medical schools, and are not necessarily reflected in college grades or in tests of "aptitude" (2). In the curriculum of the medical schools it will become increasingly important to cultivate in the students their natural humanitarian tendencies and their interest in the general welfare of their fellow man The physician's role in his community has broadened More and more he is concerned with the personal problems of his patients, their attitudes and aspirations Talking with his patient the physician finds that these matters are highly pertinent to health and disease

The precise manner in which the physician greets his patient is a matter of personal preference However, many physicians in their desire not to be austere or forbidding may assume a casual or jaunty air with cigarette in hand and feet up. Such exaggerated and unbecoming informality is usually ineffective, if indeed not actually disconcerting to a patient who hopes his problems will be taken seriously

HISTORY TAKING

In the physician's first contact with the patient he usually "takes a history", and thus sets the stage for all subsequent talking with the patient.

patients throughout the doctor's practicing career. Illustrative examples from actual student histories are quoted below:*

A young man who complained of epigastric discomfort with occasional nausea described his symptoms as follows:

Patient: I left the house feeling perfectly all right, I live in Queens, I get on at 90th Street Station, and before I reached Woodside, on the local, I began to feel kind of peculiar feeling in the stomach, no actual pain, but I might say nauseous . . .

Student: Where did you feel that?

Patient. In through there—and . . .

Student: And you felt sick at your stomach?

Patient: Yes And I get off at Queens Plaza and go across the platform to the BMT, which takes me to Lexington Avenue; it goes down into the subway. Well, when I got down underground, the perspiration just poured off me as if I was under a shower . . .

Student: This was in the evening?

Patient. No, in the morning.

Student. In the morning Were the subways crowded?

Patient: Oh, yes I was standing up .

Student: And you had a sick feeling right there?

. . . .

Student: You were coming back from what, then, shopping?

Patient: I was going to work

Student: You were going to work What kind of work do you do now?

Patient. I'm with the New York City Housing Authority .

Student. What kind of work?

Patient: Stenography and typing

Student: Stenography and typing

Patient. And . . . I ordinarily change at Lexington Avenue, and I got off there I was afraid I was going to faint, although I have never fainted in my life Things started to get grey and black, I just hung on, to this bar .

Student: This wasn't pain?

Patient Well, combined with I think the perspiration, I believe, started after the pain in the .

Student. Yes, but was it pain, or was it an uncomfortable feeling?

Patient It wasn't a sharp pain, it was just a well, I don't know . it was kind of a .

Student Dull?

Patient digestive pain, that's what I think it .

Student: Yes, but did it feel dull, was it *aching*?

Patient Yes, it was a dull pain, it wasn't a sharp pain.

Student It was dull Was it more discomfort or pain?

Patient Discomfort

---

* This material was gathered in collaboration with Doctors John T Flynn, Fred Kern, and Thomas P. Almy and is reprinted by kind permission from the Journal of Medical Education.

a social conversation with the student interrupting the patient now and then to utter words of sympathy or approbation without gathering useful data.

e. Often evident was lack of taste and discretion on the part of the student in phrasing questions and in attempting to deal with the patient's natural reticence in discussing personal matters. An off-hand approach or an introductory remark such as, "Now I am going to ask about your personal life" often put the patient on guard unnecessarily so that later questions yielded only euphemisms or actual misinformation. The sensibilities of some persons were offended by such questions as "How's the sex life?" or "Did you ever have gonorrhea or syphilis?" Some questions were phrased in a fashion entirely unsuited to the patient; for example, a candidate for the doctorate in a scientific field was questioned about "making water" rather than about "urinating". Again, a 14-year-old slow-witted schoolboy was asked "Do you repress resentment?"

Errors of Omission

This group commonly related to oversights on the student's part in not following through on obvious or possible leads offered by the patient.

a When important leads were provided by the patient's spontaneous remarks or in the manner in which he answered questions, the students frequently failed to follow them up. Often an event was carefully noted by the student but its possible implications for the health or emotional state of the patient were ignored Similarly, the student often failed to ascertain the results of previous diagnostic and therapeutic procedures

b Difficulties also arose from insufficient awareness of the individual nature of the patient and his limitations as a witness The reliability of the patient's observations was often too easily accepted A patient's estimate that an attack of syncope had lasted "ten minutes" was recorded without question, and on several occasions the patient's statement that he had never had tarry stools was not doubted until inquiry by the instructor revealed that the patient never looked at his stools The student sometimes accepted the patient's definition of an episode as a "cold" or a "heart attack" without weighing the evidence in support of these conclusions

c Often the student's preoccupation with charting procedures lost him valuable leads in the tone and quality of the patient's voice, slips of the tongue, misinterpretations, contradictory statements Similarly, obvious signs of disturbance in the patient were often missed, such as facial expressions of emotion relating to certain questions, flushing, tears, sweating, or changes in respiration

Defects in the technique of communication such as those described can form the basis of habits which may hamper effective communication with

A 63-year-old Irish-born spinster was describing joint pain in her hands and wrists.

Student. And you started to feel tired at that time?
Patient: Well, I had to go *to Boston, to a funeral* . .
Student: At the same time that you had pain in the back?
Patient. No, at the same time that I had pain here, at the same time that I had pain in the hand, *my sister was sick* . . .
Student· Then the next thing you got was pain in the hand?
Patient· In the hand . .
Student How long ago was that?
Patient. Well, I'll tell you now. Exactly that must be about six weeks
Student. Six weeks ago you first had pain in your left hand Where exactly in your hand was it?
Patient· There . . . and here I just can't catch anything with it.
Student Now are those pains both sharp and dull, or are they both dull? Aching?
Patient· I don't know The point that worries me—
Student· Now just one second, I want to write that down. That time . . . when you had the pains in this hand over here, did you have pains in your wrist, too? Were your wrists stiff at that time—was it difficult to move the hand?
Patient I tell you, doctor, I think . I haven't used this hand . . . as I told you, *my sister's husband died and she was sick, and I took care of her for a whole week and I think that is what brought this whole thing on me* Now she's well, she is taking care of me I get very tired
Student. I want to ask you some more about this hand Was it swollen at that time? Was it red or swollen?
Patient Not a bit, no change in it You see it was swollen, there, you see that
Student It was swollen right in that area, not in any other part? But it has never been red or inflamed?
Patient No, not at all, I could wash my hands and it would never even bother me
Student Is this swollen, your hand?
Patient You see, right in here Can't you see that it is swollen?
Student Yes, I can see that Can you move your fingers?
Patient Sure, well I can't very well When I have this pain here, in that hand, I can't sew
Student That is a constant pain? Never goes away?
Patient Yes, [illegible]

Student Then you got what?
Patient Here, in the shoulder, and here
Student How long ago was it that you got the pain in that shoulder?
Patient It must be five weeks, because *he has been dead a month now*
Student Five weeks ago That is the same sort of pain, an aching .
Patient A wicked pain
At this point the instructor broke in and asked· You said you were very tired, is that right?
Patient I was tired, I know the reason I was tired, *I told you I had to go to Boston to a funeral, my sister was sick I came home and nursed her for a week* She wasn't able to go to her husband's funeral I had to nurse her for a week, she had a bad cold,

Student. Yes Did it move up into your chest or down into your lower abdomen?
Patient. No

Comment: About the only objective accomplished by this staccato rhythm of questioning was the interruption of the patient's account of his symptoms which might have turned out to be coherent and informative.

One of the students who gave free rein to the patient to develop his own story collected considerable pertinent data in a relatively short time His patient, a 30-year-old salesman, had complaints similar to those of the first patient.

Student. Tell me a little bit about what brings you here

Patient: Actually, it is my stomach. I have been suffering with my stomach on and off since I was in the Army During the past 5 years I went to the doctor, once, a couple of years back, I had a check-up, went on a diet and he told me to gain some weight, but I never did I suffer indigestion quite a bit, I don't gain any weight; I feel tired and run down, so I thought I had better come down and see if there was anything wrong

Student You have been to a number of doctors—in the past five years?

Patient· I have I went to my own family doctor, and he recommended that I go to a specialist for some x-rays, so I went through a set of x-rays and they didn't find anything at that time, but that was about three years ago

Student· The last time you were well was five years ago?

Patient I think it was even more than that, because all the time I was in the Army I never ate right, when all the other guys were gaining weight and eating like horses, I never ate I never could eat breakfast and I never felt right

Student· Can you tell me then the last time you were well, as far as your stomach is concerned?

Patient I think it was back in 1941 Well, even as a youngster I had a weak stomach, I mean, it was easily upset I'd get faint very easily, if I went for a needle or a shot or a Wassermann, I'd get faint and things would upset my stomach, I'd get that sick, nauseous feeling very easily, you know Now that I think back, I always had a weak stomach

Comment· In these few minutes, the patient has had the satisfaction of speaking as much as he could or wished to about his illness Without any pressure from the physician it has become quickly apparent that he is dealing with a life-long pattern of gastrointestinal symptoms in a 30-year-old man with childish mannerisms of speech and tone of voice, and that other areas of performance will need attention beside the examination of the gastrointestinal tract.

The following is an example of disregard of an important lead on several separate occasions because of preoccupation with the mechanics of charting and with minute details of the patient's arthritic symptoms Finally, near the end of the interview a single question by the instructor brought out the pertinent material.

Student: Are you well liked by them?
Patient: I think so
Student: Are you an easy mixer? Do you get along well with people?
Patient: Well, fairly easily.
Student: Are you rather easy going?
Patient: No, I am a worrier, I worry a lot, I think
Student: Do little things upset you?
Patient. Well . . sometimes . . . they do, yes. It depends somewhat the strain I am under.
Student: Do you ever get resentful or mad at people?
Patient: Not . . usually.
Student Do you get angry easily or not so easily?
Patient Well I am sort of short tempered, I guess.
Student Are you jealous of people?
Patient: No
Student. Do you keep things inside yourself? Have you always been that way?
Patient: Yes

Comment: This type of questioning is certain to fail to obtain pertinent information and yet the answers to the questions are duly recorded in the chart however empty they are Later questioning by the instructor brought out the importance of stresses in the patient's work situation to his illness

The following example of "personal history taking" illustrates a more "inquisitional" method in which the patient is assaulted by rapid-fire questions like blows from a triphammer which shatter or threaten the patient's sense of privacy, and often lead him to openly-expressed resentment. Another technical error made by this student was offering a number of alternative answers or only one answer, instead of allowing the answer to be spontaneous The patient was a 53-year-old housewife complaining of epigastric distress.

Student How old were you when you got married?
Patient Forty
Student You were forty when you got married Had you had many opportunities to get married before that? Just waiting for the right man, or what was it?
Patient I didn't care
Student You didn't care? I don't understand you didn't care You mean you had *no desire to get*
Patient I had opportunities
Student But?
Patient but I was making plenty of money at that time .
Student And to make the money was more important than to get married and start a family?
Patient I was always afraid I wouldn't get the right man
Student You were? Why?
Patient I don't know
Student What was the matter? Had you had some bad experiences in your own family?

she had a doctor every day. And then I used to get up at night, and give her some medicine, you know. I think myself I got cold, then I got this, in my arm. And her doctor told me to bathe it with epsom salts, the hand . . well, I did it, three times a night, and the pain . . . it didn't help it one bit. I got tired then, and I think this is the cause of the whole thing. I imagine, myself . . . I don't know. I got tired . . .

Instructor: Had you travelled all the way . . .

Patient: I travelled all the way, and I kept my hand up the whole time in that car, with the pain I got to Boston, then we got off, we had to take a car to the cemetery miles out again That is where they lived before and that is where their graves are So I had to go, and that is what I really think was the cause of all my troubles. But I am not going to say it. Now she is all right, she is taking care of me . . . she is much older than I am

Instructor: You are not going to say what?

Patient: I am not going to say anything about it . I mean to say, *I don't want to resent it or anything* I am not going to think . . to let her know that was the cause of it That would make her feel bad

Comment: Here the references to the sister's illness and surrounding circumstances were missed half a dozen times by the student, who passed over them to focus on various precise and probably unimportant details of the symptoms.

In the following example is illustrated another defect in the student's technique of handling the interview so that the result is dull, sterile, and unproductive. The patient was a 28-year-old stockroom clerk who suffered from alternating episodes of constipation and diarrhea. The student quizzed him vigorously but his questions led nowhere.

Patient My parents were separated when I was eleven

Student. Whom did you live with?

Patient: My mother

Student Did you get along well with your step-father or not?

Patient Well huh not really

Student Did your brothers live with you, and your mother?

Patient. One brother did

Student: Are there things you would rather do than your present job?

Patient Well . I don't know Probably there are positions I would rather have but offhand I can't think of any

Student Are you bored with your work or not?

Patient Well, it is rather routine not too much variety, the same thing every day

Student Are you a little irritated with it at times?

Patient: Well . . . I think so

Student: Do you have a lot of friends at the present time?

Patient: Yes, I do

Student. Do you enjoy the company of these people?

Patient: Yes

Student Do you get along well with them?

Patient Yes.

Patient: I don't smoke.

Student: You don't smoke. How often do you take some kind of whiskey or beer?

Patient: I haven't had a drink in over a year. I don't drink either, except once in a while to be sociable, but I haven't even done that since I have been sick.

Student: How much sleep do you get a night?

Patient: Well . . . I try to get about eight hours every night, but I don't get that.

At this point it seemed apparent to the instructor that this patient had always had a shaky relationship with the opposite sex, and that facts about her relationship to her husband might be pertinent to her colitis. In an attempt to preserve continuity in the questioning so that the patient would not feel threatened by a sudden change of topic, he asked: Has your husband been drinking at all?

Patient: He doesn't drink either, nor smoke. [A long pause ensued here during which the patient seemed to be preparing some further statement about her husband.] My husband is at least ten years older than me, and that may explain some of our differences. I say, at least 10 years older, because, when we first met, he told me that he was ten years older, but since, I have had reason to believe that he is more than 10 years older than I am. I don't know whether that means anything, except that we don't think alike on many things . . . and he clings to very, very old-fashioned ideas about almost everything and he has something to say about everything, including things that I don't think concern him . . . like things about the house, furniture, curtains, or anything. He will come in the kitchen . . . you know, something I am cooking . . . well, he thinks he knows more about cooking than I do. He has something to say about everything.

Instructor. How much do you think these things have to do with your health?

Patient: Well, I think that must have quite a bit to do with it, I hate to say it [began to cry at this point] but in my heart I feel . . . that . . . I don't like . . . I don't like to break up my marriage . . . but I realize that . . . my husband was not the person . . . I should have married . . . it was only about six months after we were married that he struck me . . . just in a fit of temper over some silly little thing that didn't concern him.

Comment: Often by carefully watching the patient it is possible to tell that a single question is all that may be needed to bring out relevant material, but preoccupation with charting and haste to get to the next question may interfere with opportunities for such observations.

The first objectives in history taking may be epitomized as follows:

1. To make the patient feel comfortable and secure.
2. To enable him to trust the doctor's skill and judgment, his respect for confidential material and his personal motivations.
3. To make it easy for the patient to talk freely and frankly.
4. To direct the discussion along the most pertinent and profitable lines for diagnosis and treatment.

To achieve these objectives, time and patience are required, as well as dignity and sympathy of manner. Lack of time is the excuse most frequently offered by physicians for failure to understand their patients. Time is not everything. A definite skill is required. Many physicians who spend a good deal of time talking to the patient get very little pertinent information and may learn very little about the patient as a person.

Patient: No I saw a few other people, friends of mine . . .
Student. Yes
Patient: So I figured "take it easy", plenty of time.
Student Did you have a lot of boy friends?
Patient: . . Yes
Student: How old was your husband? Was he older than you or younger . . .
Patient: He was in his forties
Student: In his late forties? How much older was your husband than you?
Patient: Five.
Student: Five Where did you meet him?
Patient. Do I have to tell you all that?
Student No, you don't have to It is perfectly all right. I don't care.

Comment: His final statement reflected more clearly than he realized the attitude of this student in dealing with his patient. It is unlikely that the physician who doesn't care will very often be able to create a situation suitable for talking with his patient.

Occasionally the student's line of questioning, oblivious as it was to the needs of the patient, so antagonized the latter as to preclude any further gathering of information.

A "routine" personal history on an extremely tense 48-year-old white married woman with complaint of frequent loose stools illustrates not only indifference and gaucherie as above but cursoriness and lack of curiosity on the part of the student:

Patient: I just got married, it must be about 4½ years ago
Student Had you ever considered marriage before that?
Patient Oh yes, a couple of times before that
Student And you hadn't undertaken that, however?
Patient: No
Student. Was there any reason?
Patient: No oh, there are reasons This boy I went out with was a neighbor and well, he just couldn't settle down to saving or get to the point where we were getting anywhere, so we finally broke off after going around with him for about 10 years I just went out with a couple of men in my life for any length of time
Student I understand Do you eat three meals a day?
Patient Three?
Student Yes Do you usually have some meat or fish every day?
Patient Yes
Student: Fresh vegetables, fresh fruit?
Patient I won't say that I eat fresh fruit every day We usually have juice in the morning, usually grapefruit juice in
Student All right How frequently do you have eggs?
Patient Well . . ah I used to have an egg almost every morning, that was before I was married, well my husband doesn't care for them, so I don't bother about them for myself But in the past few months since I haven't been feeling well, I have been eating soft boiled eggs Now they sort of well, I don't know whether they disagree with me, I have sort of taken a dislike to them
Student: How many cigarettes do you smoke a day?

Patient: I don't smoke.

Student: You don't smoke. How often do you take some kind of whiskey or beer?

Patient. I haven't had a drink in over a year. I don't drink either, except once in a while to be sociable, but I haven't even done that since I have been sick.

Student: How much sleep do you get a night?

Patient: Well . . . I try to get about eight hours every night, but I don't get that.

At this point it seemed apparent to the instructor that this patient had always had a shaky relationship with the opposite sex, and that facts about her relationship to her husband might be pertinent to her colitis. In an attempt to preserve continuity in the questioning so that the patient would not feel threatened by a sudden change of topic, he asked Has your husband been drinking at all?

Patient He doesn't drink either, nor smoke [A long pause ensued here during which the patient seemed to be preparing some further statement about her husband] My husband is at least ten years older than me, and that may explain some of our differences I say, at least 10 years older, because, when we first met, he told me that he was ten years older, but since, I have had reason to believe that he is more than 10 years older than I am I don't know whether that means anything, except that we don't think alike on many things . . and he clings to very, very old-fashioned ideas about almost everything and he has something to say about everything, including things that I don't think concern him like things about the house, furniture, curtains, or anything. He will come in the kitchen you know, something I am cooking well, he thinks he knows more about cooking than I do He has something to say about everything.

Instructor: How much do you think these things have to do with your health?

Patient Well, I think that must have quite a bit to do with it, I hate to say it [began to cry at this point] but in my heart I feel . that I don't like . I don't like to break up my marriage . but I realize that my husband was not the person I should have married it was only about six months after we were married that he struck me just in a fit of temper over some silly little thing that didn't concern him

Comment Often by carefully watching the patient it is possible to tell that a single question is all that may be needed to bring out relevant material, but preoccupation with charting and haste to get to the next question may interfere with opportunities for such observations

The first objectives in history taking may be epitomized as follows:

1. To make the patient feel comfortable and secure
2. To enable him to trust the doctor's skill and judgment, his respect for confidential material and his personal motivations
3. To make it easy for the patient to talk freely and frankly
4. To direct the discussion along the most pertinent and profitable lines for diagnosis and treatment

To achieve these objectives, time and patience are required, as well as dignity and sympathy of manner Lack of time is the excuse most frequently offered by physicians for failure to understand their patients. Time is not everything. A definite skill is required Many physicians who spend a good deal of time talking to the patient get very little pertinent information and may learn very little about the patient as a person.

LEARNING MORE ABOUT THE PATIENT AS A PERSON

It is usually easier to gather data regarding the personal lives of patients if questions are asked as they seem pertinent in the context of "present illness", "past history", or "family history" rather than in the separate category, "personal history".

The general method of questioning must be such as to provoke informative answers Often when a physician is unable to elicit useful personal data, the difficulty appears to lie largely in the way questions are asked. "What kinds of things were you punished for when you were a child?" is a much better question than "Did your parents punish you very much?" Questions which invite euphemistic or platitudinous replies, which gloss over the problem, must be avoided in favor of questions which set the patient to thinking about himself

A most common, and not very profitable, practice among physicians is to ask a few direct questions to cover the most likely areas of conflict, as, for example:

"How are things at home?"
"Does your wife nag?"
"Do the children annoy you?"
"Have you and your wife made a good sexual adjustment?"
"Do you have financial worries?"
"How are things at the job?"
"Is your boss unreasonable?"
"Do you get on well with your fellow workers?"
"Are there any worries that you can think of?"

Such questions may elicit the required information in a few patients who already have a considerable awareness of their problems and who are not too diffident to discuss them Most patients, however, will make benign part-answers to these questions and then leave the physician at a loss to know what to do next How to proceed from here? It is best to avoid such blind alleys in the first place and approach the patient in an altogether different way. One simple, and usually effective, way to sift out the possibly relevant conflicts, even when the patient is unaware of them, is to elicit a brief panorama of the patient's background, aspirations and life experiences Such a story requires 5 to 10 minutes to tell and it offers the physician the opportunity to suspect from his own experience and knowledge of life and people wherein fulfillment or satisfactions may be lacking Furthermore, merely by telling his story the patient may unconsciously provide strong and reliable hints of inner conflict The physician now has something to go on and can direct his inquiry into the most likely channels. He is not "flying blind" as in the following example, a verbatim excerpt from a physician-patient interview

The patient was a 40-year-old dentist of Russian Jewish parentage with

peptic ulcer and migraine headaches. He was welcomed into the office by the physician and after he had described in detail his symptoms and past illnesses and after a physical examination which failed to turn up any abnormality, the physician sat down with him a bit uneasily and said:

Doctor: As you probably know, a good many physicians connect ulcer and migraine with emotional problems, so I think it would be well if we reviewed the situation together a little bit Of course, you realize that anything you say to me is entirely confidential.

Patient: Yes, I know that, Doctor, and I've given this thing considerable thought myself I thought there might be some nervousness or emotional troubles, but for the life of me I can't find a thing I've got a fine wife, two lovely children, an excellent practice and a house of my own We have no in-law troubles and I should be the happiest man in the world

Doctor. Well, tell me some of the details Is there any disturbance at home? Do the children get on your nerves?

This line and several others were pursued with negative results and finally the physician asked:

Well, what about the office? Is there anything down at the office which isn't going just right? The landlord, your secretary, the place itself? Are you making as much in practice as you hoped you would?

Patient Really, Doctor, I'm doing better than I ever expected I can't complain of my practice in any respect It's as good as any dentist I know

This physician was approaching the problem in an earnest and systematic way and was getting nowhere for the following reasons:

1 Because too much was made of the fact that the physician was now going to "launch into a discussion" of his patient's personality adjustment, the patient was put on the defensive. The data could have been elicited along with the rest of the history and prior to physical examination

2. The questions were asked with an implication of possible points of conflict without the physician really knowing anything about the patient, his hopes and fears and aspirations On the other hand, he could have suspected possible areas of trouble from a superficial knowledge of the cultural and social setting from which this individual came

A second physician, to whom this patient was referred, was able to bring into focus one factor of significant personal conflict in an interview which lasted the same length of time as that of the first physician

It is important to note that the patient was trying to be honest with both physicians He had truthfully denied the possible sources of worry suggested by the first because the first line of questioning did not lead him to see himself the way that of the second physician did The differences may seem subtle but they are fundamental to success in talking with a patient An excerpt of the interview with the second physician follows

Doctor Are you doing with your life what you want to do?

Patient (long pause) I can't be definite about that, Doctor I *believe* I am I *think* I'm content

Doctor: What kind of a practice did you plan for yourself when you first started out?

Patient: Well, that might be a source of discontent to myself. I have a fairly busy practice and I don't think it's—perhaps the quality isn't there. And that might be a source of discontent in my own mind.

Doctor: You mean that it's too routine?

Patient. Well, I think perhaps seeing too many patients and you can't be as selective in your work, your quality is perhaps not as good as you'd like it to be. Now, I might be theorizing there.

Doctor Well, go ahead

Patient. Those are possibilities that loom in my mind. I, I, uh, I have a busy practice, but perhaps it isn't a happy practice, though, in my own mind

Doctor: What would you—suppose all stops were out—what would you like to be doing?

Patient: Just what you're doing now. I've often thought I'd like to be affiliated with a University. I know I enjoyed my residency, my internship and residency *immensely*. I stayed on for about 5 years and I got a big bang out of that I enjoyed it *immensely* I was very happy there.

Doctor When was this?

Patient: In 1937 Graduated, then interned I stayed on as resident.

Doctor And then did you think at the time of continuing in full time work, teaching?

Patient. Well, I opened an office not far from the hospital so that I could retain my position at the institution

Doctor: You still do that?

Patient No I gave that up I guess somehow or other I got into a—I just got into a routine, we get ourselves married and we get into that sort of a swing, as it were

Doctor The need for better income?

Patient I would imagine so

Here the patient's manner was grim and dejected and it was evident that an important area of conflict had been discovered The discussion moved logically to the question of his wife's financial demands and her social ambitions Soon the story of feeling caught in a treadmill, trying to supply the demands of an avid and ambitious wife came out in clear relief. He wept during much of this discussion, but at the end he felt much relieved at having been able to discuss matters which were so close to him that he was hardly aware of them

In this case the physician had succeeded in talking with the patient because his questions were brief and sympathetic and provocative of thought. The patient was induced to tell about himself, in his own words and thereby provided the examiner with leads as to where his trouble lay. These are the "presystolic rumbles" of inner conflicts

Typical ways in which patients unconsciously betray inner conflicts are by unaccountable memory defects for past events, contradictory statements, mention of a person or subject out of the direct line of the story, damning with faint praise and "blocking", or suddenly changing the sub-

ject before the previous thought has been developed. These and other signs in the interview, including gestures, postures, facial expressions and tone of voice give important information to the attentive observer about what makes his patient "tick". They are among the important data elicited by the skillful physician in talking with his patient.

DEALING WITH UNCONSCIOUS MATERIAL

Waiving for the moment the need to subscribe to any fixed doctrine or to interpret findings in the light of a formula concerning the maturation process, I think we can all agree that activities go on within us of which we are unaware or only partially aware This generalization applies, of course, to such visceral functions as heart action, digestion, and spermatogenesis. These unconscious effector activities are organized by an equally unconscious process in the brain. Receptor activities too may also be unconscious We have a strong body of evidence to this effect. Anyone who has used a monocular microscope leaving the non-operative eye open can be easily convinced of this Cross eyes provide another example. The cross-eyed subject's visual receptor apparatus may be intact but he is conscious only of seeing in one direction. The chain of unconscious mental activity is completed when we link up unconscious receptor processes with the unconscious integrative activity of interpretation and mentation so that effector responses or reactions can be formulated and executed

Thus unconscious receptor activity may lead to more or less observable changes such as word or thought associations, dreams, slips of the tongue and special behavior, gestures, facial expressions, and other skeletal muscle and visceral changes By the same route conscious receptor activity may, of course, lead to this chain of events but it is far more obvious.

The distinction between conscious and unconscious depends upon a quantitative factor, namely the subject's degree of awareness This may be great, small or apparently lacking and it may fluctuate, being absent one day and present the next. The "unconscious" is therefore an arbitrary categorical concept which applies when awareness is small or lacking but it does not necessarily imply a qualitative difference in the biologic process

Thus it is not surprising to find that physiologic changes occur during conflict characterized by all degrees of awareness. Often suppression or repression seemed to intensify or prolong physiologic disturbances but unconscious mental activity has not been found to be the determining factor for the occurrence of such bodily reactions (4)

Access to the unconscious may be had by a variety of maneuvers. The currently popular Freudian technique of "free association" is a laborious and time-consuming one but may be the most thorough. It usually involves having the patient come to the doctor's office 5 days a week for one, two

Doctor: What kind of a practice did you plan for yourself when you first started out?

Patient: Well, that might be a source of discontent to myself. I have a fairly busy practice and I don't think it's—perhaps the quality isn't there. And that might be a source of discontent in my own mind.

Doctor: You mean that it's too routine?

Patient: Well, I think perhaps seeing too many patients and you can't be as selective in your work; your quality is perhaps not as good as you'd like it to be. Now, I might be theorizing there

Doctor: Well, go ahead.

Patient: Those are possibilities that loom in my mind. I, I, uh, I have a busy practice, but perhaps it isn't a happy practice, though, in my own mind.

Doctor: What would you—suppose all stops were out—what would you like to be doing?

Patient. Just what you're doing now. I've often thought I'd like to be affiliated with a University. I know I enjoyed my residency, my internship and residency *immensely* I stayed on for about 5 years and I got a big bang out of that. I enjoyed it *immensely* I was very happy there

Doctor: When was this?

Patient: In 1937 Graduated, then interned I stayed on as resident.

Doctor: And then did you think at the time of continuing in full time work, teaching?

Patient. Well, I opened an office not far from the hospital so that I could retain my position at the institution

Doctor: You still do that?

Patient No I gave that up I guess somehow or other I got into a—I just got into a routine, we get ourselves married and we get into that sort of a swing, as it were.

Doctor The need for better income?

Patient: I would imagine so

Here the patient's manner was grim and dejected and it was evident that an important area of conflict had been discovered. The discussion moved logically to the question of his wife's financial demands and her social ambitions. Soon the story of feeling caught in a treadmill, trying to supply the demands of an avid and ambitious wife came out in clear relief. He wept during much of this discussion, but at the end he felt much relieved at having been able to discuss matters which were so close to him that he was hardly aware of them

In this case the physician had succeeded in talking with the patient because his questions were brief and sympathetic and provocative of thought. The patient was induced to tell about himself, in his own words and thereby provided the examiner with leads as to where his trouble lay These are the "presystolic rumbles" of inner conflicts

Typical ways in which patients unconsciously betray inner conflicts are by unaccountable memory defects for past events, contradictory statements, mention of a person or subject out of the direct line of the story, damning with faint praise and "blocking", or suddenly changing the sub-

conflict within herself about these strivings. These remarks viewed against her statements about her close relationship with her father suggested that her "deep understanding" of her father was really a matter of striving to please him.

These implications were confirmed four sessions later when spontaneously she stated, "Daddy wanted to be internationally known—he was not. He was terribly frustrated. I wanted, in some way, to compensate for his insignificance, as he interpreted it, by doing something big myself. I swear that's the truth . . . *I've never thought of that before*, but I'm sure of it now. I think I wanted to fulfill a lack in Daddy's life . . . There were so many disappointments in his life . . . He was a very unhappy, inwardly person and I had this tremendous desire, as *I look at it now, which I never thought of before*—but I wanted to, as I said, to in some way compensate for Daddy's lack by being able to accomplish something myself."

Later after she had been asked to think further about her father she said, "But in *going back and analyzing, I've remembered things that I'd forgotten* and I wished that I'd still forgotten Daddy was cruel to animals occasionally and I can remember bursting into tears and sobbing when he kicked Andy, our little yellow dog, a fox terrier I can still remember his ill humor, exactly like Jack's, in the early morning; I can remember his anti-social attitude, which was a defense mechanism on his part because he felt that he had not gained the prestige that he deserved and, therefore, he was 'agin' all men. *I remembered all of that and a great deal more which I had hidden completely because of my idolatry for him as*—I built him up to be or as he was; that I don't know."

These passages not only indicate the reliability of indirect signs of conflict in early interviews but also illustrate how the physician without special training in psychoanalysis can with skill and patience elicit important data from the unconscious.

DANGER SIGNALS

Often physicians are reluctant to explore very vigorously their patients' unconscious thoughts or wishes for fear of doing harm Such fears are unnecessary if one is alert to the early signs of "overexploration". They are just as recognizable as are the first indications of digitalis intoxication The first and most likely ones are the premonitory signs of serious depression. One of these is a stubborn hopelessness of attitude in the face of facts that seem encouraging to the physician. Another is a persistent attitude of self blame, especially in relation to actions which are not logically blameworthy. Other signs include the appearance of extreme insomnia, agitation, delusions or difficulty in organizing thoughts When such signs occur one should of course discontinue further exploration for a time It is not wise, however,

or more years. He lies supine on a couch facing away from his physician and says aloud whatever comes into his mind. Occasionally the analyst guides the patient's remarks into general topics of current events on the one hand or old recollections on the other. When this method is successful the patient ultimately begins to recall and to say things of which he had been altogether unaware. When these data allow a better understanding of the patient's emotional development, attitudes and behavior, important conflicts may be resolved and symptoms dispelled The reciting of dreams is also a part of the psychoanalytic procedure. The patient then "free associates" from the various details in the dream so that an interpretation can be achieved which may elucidate further the unconscious processes.

Simpler indicators of the patient's unconscious mental processes are some which have already been mentioned, slips of the tongue, evasions, incidental remarks and unaccountable loss of memory for events. So are implications of general appearance, dress, manner of speaking, posture, and reactions in the company of others. Skill in interpreting these signs, like that required for interpreting heart sounds, is based on an awareness of their importance, painstaking care in observation, and practice.

An example of a patient giving inadvertently important unconscious data is provided by a young woman who told of her father's death in these words: "I was the only person Daddy recognized. He died within a few hours after I got there He went into a coma soon after I got there. But he did come out of it long enough to recognize *me*. He hadn't known Mother or anyone else, but Daddy and I could speak without words—there was a great thought wave there, always has been—is still, as far as that's concerned and, of course, I know that Daddy's had a tremendous amount to do with everything—Daddy was one of the most wonderful people that ever lived in this world, a brilliant mind, so well informed; he was a walking encyclopedia " The description of her "intimacy" with her father is informative chiefly because it doesn't reflect any real intimacy After speaking of being extremely close to him she describes him in terms of intellectual accomplishment, rather than in terms of warmth, understanding, and generosity.

Earlier in telling about her school days she had said, "I don't think that Mother and Daddy ever pushed me They were very quietly happy about any honor that I got and when I graduated magna cum laude they were glad; they were glad when I made the honor roll every term, but they never expressed themselves like, 'This is perfectly wonderful' or 'You should do it' or 'Go ahead and do it.' I just drifted through life taking everything in my stride." Such a strong need to excel rarely, if ever, occurs spontaneously. The exaggerated smoothness implied in "They were quietly happy" and "I just drifted" were considered by the physician to indicate serious

to have a penetrating knowledge of psychology and a lively interest in what makes people behave as they do.

Fifty years ago a doctor could do an effective job with a lot less than this, but that was the day when the hospital wards were filled with typhoid fever. People were dying in large numbers of pneumonia and diphtheria. Today in the wards of the hospitals such diseases as high blood pressure, peptic ulcer and diabetes predominate. There is much evidence to suggest that these are diseases of the wear and tear of living. We have no drugs or formulae to cure or prevent these diseases. Antibiotics and hygienic living have diminished the prevalence of infectious diseases, but they have not simplified the doctor's job. They have only altered the requirements for a good effective doctor. Modern doctors should be able to detect evidence of strain in patients who appear to be perfectly adjusted and may themselves be totally unaware of serious emotional problems. Moreover, the good doctor should be able to treat most of these patients.

The importance of the psychiatric approach in understanding a medical problem and treating it properly is graphically illustrated by the case of a young woman of 33 whose intense pain in the flank had been attributed to a floating kidney. Several specialists had treated her but nothing they did seemed to help. She was a cheerful, friendly, and attractive girl who seemed very well balanced indeed. She did have serious inner conflicts, however, which ultimately were found to be responsible for her pain In the early interviews with her, although she praised her husband to the skies, the physician got the impression that he was more of an "eagle scout" than a husband. Finally, it became evident that her pains came on most intensely on the mornings after intercourse with him Later, working with both her and her husband using the methods already outlined, it was possible for the non-psychiatric physician to help her resolve some fundamental problems Her pains disappeared completely and she has remained well for three years. Her difficulty was not an uncommon one Every day such patients pose serious problems to surgeons and physicians who all too commonly attempt to solve them by laparotomy without ever really talking with the patient.

Psychiatry is as important in general medical practice as is the stethoscope. The psychiatrist himself has a special interest in this field and the background of an expert, but the fundamentals of psychiatric method must be practiced by every medical man today if he is to care adequately for his patients.

TURNING TALK INTO TREATMENT

Every contact with the patient has implications for treatment and therefore may have a favorable or unfavorable effect on the course of illness. The

to drop the topic under discussion like a hot potato, but rather one should turn the discussion vigorously into an encouraging vein, complimenting the patient on his handling of the problem and generally offering him strong reassurance and support.

A second complication of overexploration is the development of panic reactions. These can be avoided if the physician is alert for early signs of fear in his patient, either as manifest in appearance, mannerisms, pulse, breathing or by an almost incongruous failure to acknowledge an obvious connection between events and his reactions When a patient is either over-reacting or protecting himself in this fashion, it is best to change the subject to neutral topics and continue for a time with an attentive supporting attitude deferring for the time being any further exploration.

A third complication of overexploration may be disorganization of thought and talk as occurs in a schizophrenic reaction. Earliest signs of this serious difficulty are striking aberrations in logic, non-sequetur responses and inability to follow out a train of thought. Sometimes the patient may make gratuitous statements which don't "hang together". Here again, exploration must stop for a time and be replaced by strong support and reassurance.

The development of subtle evidences of impending depression, panic, or disorganization may seem to provide a compelling reason to refer the patient to a psychiatrist. This is, of course, appropriate but must be accomplished skillfully and gradually and without any implication of rejection of the patient by the original physician It might be fitting here to suggest how much of psychiatry the internist or general physician requires in order for him *to function effectively in talking with his patient.*

HOW MUCH KNOWLEDGE OF PSYCHIATRY IS NECESSARY?

He needs to know at least as much about emotions as he does about drugs or microbes How he can get such knowledge is a question that is not as easy to answer A large proportion of doctors practicing medicine at the present time went to medical schools where little or no psychiatry was taught to the students. That was doubtless a handicap but it did not rob these men of the opportunity of learning psychiatry In fact, some of them have actually become psychiatrists A major portion of every doctor's fund of knowledge is learned years after graduation from medical school by virtue of his own curiosity and powers of observation He learns what he thinks is important to him. Comparatively few doctors have been aware of how important a keen understanding of the patient as a person really is to them (5)

A naturally humanitarian attitude and an interest in people have always been considered to be important ingredients of the good doctor Only in the last few years, however, has it become recognized as desirable for the doctor

to have a penetrating knowledge of psychology and a lively interest in what makes people behave as they do.

Fifty years ago a doctor could do an effective job with a lot less than this, but that was the day when the hospital wards were filled with typhoid fever. People were dying in large numbers of pneumonia and diphtheria. Today in the wards of the hospitals such diseases as high blood pressure, peptic ulcer and diabetes predominate. There is much evidence to suggest that these are diseases of the wear and tear of living. We have no drugs or formulae to cure or prevent these diseases. Antibiotics and hygienic living have diminished the prevalence of infectious diseases, but they have not simplified the doctor's job. They have only altered the requirements for a good effective doctor. Modern doctors should be able to detect evidence of strain in patients who appear to be perfectly adjusted and may themselves be totally unaware of serious emotional problems Moreover, the good doctor should be able to treat most of these patients.

The importance of the psychiatric approach in understanding a medical problem and treating it properly is graphically illustrated by the case of a young woman of 33 whose intense pain in the flank had been attributed to a floating kidney. Several specialists had treated her but nothing they did seemed to help. She was a cheerful, friendly, and attractive girl who seemed very well balanced indeed. She did have serious inner conflicts, however, which ultimately were found to be responsible for her pain. In the early interviews with her, although she praised her husband to the skies, the physician got the impression that he was more of an "eagle scout" than a husband Finally, it became evident that her pains came on most intensely on the mornings after intercourse with him. Later, working with both her and her husband using the methods already outlined, it was possible for the non-psychiatric physician to help her resolve some fundamental problems Her pains disappeared completely and she has remained well for three years. Her difficulty was not an uncommon one Every day such patients pose serious problems to surgeons and physicians who all too commonly attempt to solve them by laparotomy without ever really talking with the patient.

Psychiatry is as important in general medical practice as is the stethoscope. The psychiatrist himself has a special interest in this field and the background of an expert, but the fundamentals of psychiatric method must be practiced by every medical man today if he is to care adequately for his patients.

TURNING TALK INTO TREATMENT

Every contact with the patient has implications for treatment and therefore may have a favorable or unfavorable effect on the course of illness. The

extraordinary power of words or thoughts communicated without words is manifest in the widespread physiologic effects which have been shown to follow the administration of placebos (6). Appropriately administered, ipecac has relieved nausea and lactose capsules have induced a skin rash typical of drug sensitivity (7).

In treatment of any disorder the attitude of the physician will weigh heavily in the balance. A pharmacologic agent or a manipulative procedure administered with enthusiasm and with hope has its greatest chance of success. Bed rest, diet and other therapeutic rituals owe a part of their benefit to the quality of the communication from physician to patient. When "talking with the patient" is deliberately undertaken as therapy, there are several measures the physician might take in an effort to help the patient to a more constructive and less costly adjustment. The simplest and most applicable of these possibilities is for the physician to display an earnest and sympathetic interest in the patient as a person, to encourage him to confide whatever doubts and conflicts he may be able to discuss and to listen without implication of judgment or censure. The reassurance and emotional support for the patient which stem from this attitude on the part of the physician have been shown to be the most powerful psychotherapeutic tool available The physician may also attempt by various means to uncover repressions and induce the patient to a reorientation of attitude toward people and events This maneuver requires special technical training. The physician who is not a technical specialist, however, may accomplish a great deal by assuming a dependable and persuasive attitude which allows the *patient vicariously to work off his bottled-up hostilities and to expiate his* guilt It is equally essential to give serious and attentive consideration to all bodily symptoms even though no explanation may be offered and no medication given for them Much damage can be done by "brushing off" or being impatient with the patient's complaints An explanation of the mechanisms responsible for symptoms is often appropriate but it is unwise to "push" them if the patient does not readily accept them Especially, it is not wise to force the patient to "insight", to induce him to acknowledge a connection between his personal conflicts and his symptoms Very often a patient will deny such a relationship and yet paradoxically will talk freely as if he were aware *of the connection This "face saving" screen is frequently* very important to the patient There is often little to be gained and much to be lost by breaking it down The physician can sometimes help in the resolution of dilemmas and mitigate the doubts and conflicts of indecision by weighting the balance indirectly by discussion Obviously it is necessary

[illegible]

proach; counselling, non-directive, suppressive and expressive in counsel-

ling, the physician actively advises the patient and helps him work out the solution to his problems. In the non-directive approach the physician acts as a sounding board for the patient thinking out loud, as it were, in an effort to arrive at a satisfactory course of action. In suppressive therapy, the emphasis is put on building the patient's confidence and on exploiting all possible sources of satisfaction in the patient's life. In this way it is hoped that the destructive effects of unconscious conflicts will be proportionately minimized. In expressive therapy the emphasis is on uncovering repressed material so that the patient can face the issues in perspective. Thus he may realize that powerful childhood conflicts need no longer apply to his scheme of life and with the help of his relationship with the therapist he may "grow up" and achieve a greater degree of emotional maturity.

None of these measures is offered in "pure culture" but the wise physician varies his emphasis with each patient, changing from time to time as the situation demands.

Effective treatment in the form of talking with the patient does not require adherence to any specific doctrine or theory. Neither does it necessarily require couches, certificates or other trappings in the doctor's office. It does, however, require a thorough knowledge of contemporary psychology, careful training, and experience in the diagnostic and therapeutic use of the interview.

Finally, the power inherent in talking with the patient calls for a physician with a wide range of personal experience, a broad view of the phenomena of health and disease and a comprehensive approach to each patient and his individual problem (8) The development of these qualities must become a subject for special emphasis in medical schools in the future Thus the modern emphasis on the "whole patient" calls for the development of a "whole doctor"

REFERENCES

1 Wolf, S and Wolff, H G Notes on a Symposium The internist as a psychiatrist Ann Int Med, 34: 212, 1951

2 Wolf, S He goes through the motions—lulling Ment Hyg, 34: 185, 1950

3 Flynn, J T, Kern, F, Almy, T P and Wolf, S Instruction in medical history-taking, the use of wire and tape recorders Jour of Medical Educ July 1952

4. Wolf, S and Glass, G B J Correlation of conscious and unconscious conflicts with changes in gastric function and structure and observations on the relation of the constituents of the gastric juice to the integrity of mucous membrane. Proc Assn Res Nerv & Ment Dis, 29: 665, 1950

5 [illegible]

6

7 Wolf, S Unpublished data

8 Wolf, S The whole doctor Curr Med Dig, 18: 31, 1951

Precordial Noises Heard at a Distance from the Chest

WILLIAM BENNETT BEAN, M.D.

INTRODUCTION

My interest in precordial noises heard by the unaided ear at a distance from the chest began in 1938 on Dr. Blankenhorn's Medical Service at the Cincinnati General Hospital when I accidentally produced such a noise in a patient in congestive heart failure whose left lung was punctured during what had begun as an ordinary thoracentesis (1). When I visited him again several hours later he had been alarmed by a noise "like a paddle wheel makes on the river"* which he could hear and "feel". When the room was quiet sounds were barely audible at the foot of the bed. They could be heard clearly two feet away The sounds were synchronous with cardiac systole. Left sided pneumothorax was demonstrated by physical signs and by x-ray, but no subcutaneous or mediastinal emphysema was found. The loud crackling noises (Hamman's sign) hurt the ears when the stethoscope was applied to the precordium All abnormal sounds had gone in two days and the patient made an uneventful recovery. This chance occurrence became "a burr that sticks in the memory" because of its close temporal association with two cases of interstitial emphysema of the lung which McGuire and I (1) published soon after Hamman had aroused interest in the dramatic clinical features of the spontaneous form of this disorder. Another circumstance which gave my interest a wider sweep was a casual comment in Osler's essay on Sir Thomas Browne (2) in which he quoted from a letter about a "woeman or mayd in Suffolk who had a julking and fluctuation in her chest heard by the standers by" (3) though this proved to be pyopneumothorax and a succussion splash (see fig. 1)

Since these events focussed my attention on noises heard at a distance from the chest I have been on the lookout for other examples This interest has been rewarded by finding several cases Medical reports of these and other kinds of unusual and alarming noises heard at a distance have been found under the most diverse titles and out-of-the-way places No cumulative index gives a clue to this sign and though my reading has been wide surely many cases have been missed Though I conceived the notion that I would review all published cases and produce an all-inclusive monograph this has not been possible without an encyclopedantic approach time-con-

* This simile and others likening the noise to "the grinding of gears", "the rustling of leaves", "crunching up newspapers" are a pleasant tribute to the fertility of folk speechways.

Sir Thomas Browne to his son Edward.

[MS. SLOAN. 1847.]

Jan. 5, [1679-80.]

DEAR SONNE,

Present my service to Sir John Churchman and his lady. Sir John is a discreet and sober person and courteous, and his lady, though shee bee somewhat hypochond. fearefull, and complayning, is a kind gentlewoman; they have been used to apply unto some one physitian in London, and not more, I thinck, except upon very great occasion. Sir John hath an estate within four miles of Thetford: unto which hee cometh every yeare about July, and returnes in October. Autumne was twelvemoneth his lady fell sick of the autumnall spurious ague, and I being then within three miles, shee sent to mee, I found her weake and dispirited, despondent, and even despayring ever to go to London agayne; butt I comforted her to some good satisfaction, though I conceave shee did butt half beleeve my predictions of her recovery in time. I knowe not where in London shee liveth; remember mee also to her daughter, who is a sober and modest young gentlewoeman; they had also a sonne or two, butt young; the answering their doubts doth give them good content. Dr. Jasper Needham hath showne himself a kind and right honest gentleman, and you may wish well unto his relations.[7] There was a woeman or mayd in Suffolk who had a julking and fluctuation in her chest and somewhat upwardly: so that when shee stood and stroked her chest it might be heard by the standers by, and I once heard it, shee dyed, as I remember, about a yeare and half after, and in her chest was found a cystis containing above a quart, as I take it, of a matter like thick whaye, of this, Dr. Fairfax, now of Woodbridg gave an account to the R. S. about seven yeare past, and it is printed.[8] There is a man in

[7] He died in the preceding November

VOL I T

FIG 1 PART OF LETTER FROM SIR THOMAS BROWNE TO HIS SON, EDWARD. From Vol 1, p 273, The Works of Sir Thomas Browne, including his unpublished correspondence and a memoir Edited by Simon Wilkin, F L S Henry G. Bohn London, 1846

mination. Most of the types of lesions and disordered functions which give rise to such sounds will be considered. While such a miscellany may seem whimsical there are useful lessons in diagnosis enabling the physician to give correct therapy, which may vary from reassuring the disturbed patient who has mediastinal emphysema to removing air from the right ventricle of a heart churning with air embolism and thus saving a life.

DEFINITION

I use the term precordial noises or sounds audible at a distance from the chest to imply unusual loudness (rather than overacute hearing) and to indicate that the heart's action is responsible for their production. This excludes wheezes and pleural friction sounds, and even ribs cracking with a loud sound (4). The quantitative aspects defy analysis since the subjective element in hearing has not been measured or compared. General impressions, when supported by substantial clinical fact, suffice for my purpose. Though the noises discussed here usually come from outside the heart, and most often are produced by impingement of the heart upon air, or air and fluid, in abnormal places, they may be produced within the heart as extremely loud murmurs or by intracardiac air Laennec (5) was of the opinion that air was required for the production of all such sounds emanating from the animal body. Borborygmus might gain a questionable place in this strange list of noises, but has been relegated to a footnote In like manner the succussion splash known to Hippocrates, a very unpleasant sound, has been excluded since it does not occur without active or passive agitation of the chest and usually has no cardiac element Alteration of structure or abnormality of function of lung, pleura, mediastinum, pericardium, alimentary canal and heart causes such noises under the peculiar and sometimes poorly understood circumstances now to be set forth

HISTORICAL NOTE

Very ancient medical records contain no trace of instances of sounds heard at a distance from the chest There is no thoracic counterpart of the borborygmal chorus of Aristophanes' *Clouds* Only with the perfection of the art and science of physical diagnosis was an interest in pulmonary and cardiac sounds and noises developed sufficiently to get cases into the record. Some of the early instances of extrathoracic noises were recorded by Laennec* in his treatise on auscultation (5) Almost every kind of condition capable of producing such noises was recognized and identified during the resurgence of physical diagnosis in the 19th century, with contributions

* [illegible] "a heart murmur perceptible at a distance" in himself during [illegible] semi-erect position and was heard shortly

about evenly divided between French, British, and German clinicians. Experimentally many types of noises have been produced by a number of investigators but their widely scattered work has not been gathered for review. I will comment on appropriate early observations in each section of the paper.

CHEST AND MEDIASTINUM

Pneumothorax

Sounds heard at a distance from the chest have been described in left-sided pneumothorax—the spontaneous, therapeutically induced and traumatic variety. An open chest wound increases the likelihood of such noises occurring after injury. This sign is well known among clinicians familiar with therapeutic pneumothorax for pulmonary tuberculosis Indeed the act of inducing pneumothorax on the left side may cause unusual noises heard by the patient, attendants and physicians. A division of cases has been made on the basis of the source of air in the thoracic cavity. Since trauma necessarily preceded therapy historically the subject is discussed in this sequence

TRAUMATIC PNEUMOTHORAX

It is by no means clear that some cases discussed under hydropneumothorax, notably those of Bricheteau (6), are not actually traumatic pneumothorax or pneumomediastinum rather than pneumopericardium which he diagnosed. Furthermore, it is very likely that some of the German cases of frightening Mühlengerausch, which are attributed to air embolism, belong in the category of traumatic pneumothorax or mediastinal emphysema Probably a different mechanism causes the sounds in left sided pneumothorax and mediastinal emphysema; either may occur without clear proof of the other On the other hand, many cases have x-ray evidence of air in both places at the same time and it cannot be denied that the two lesions tend to occur at the same time My assumption is that a different process may cause the noises in the two conditions

Morel-Lavallée (1864) reported a 27-year old coach driver who was run over by a carriage (7) His chest was bruised and several ribs were broken. Subcutaneous emphysema was noted The *bruit de moulin** was heard everywhere near the bed The sound lasted for five days, and was so troublesome at night that the man could scarcely sleep. In due course the cure was complete Although Morel-Lavallée thought the cause of the trouble

* The noise of the mill This is a very happy simile since the combination of water splashing rhythmically and a background sound of wooden machinery and running water is elegantly reproduced in miniature by the sound of pneumopericardium or pericardial knock, as anyone knows who has listened to both

was not rupture of lung but perforation of the pericardium, traumatic pneumothorax or mediastinal emphysema seems more probable.

Reynier (1880) was another early observer of the *bruit de moulin* in cases of chest injury (8). He described a noise heard at a distance in a man who fell and broke five ribs. Similar noises in which a metallic bruit, a tinkle or a systolic sound was heard were recounted in instances of pistol wounds and carriage or wagon accidents. In several cases it is not clear that the sounds were audible to the naked ear. He reported a number of studies with air injections into small animals where similar auscultatory signs had been produced. He quotes Aran as having injected air into the pericardium of animals and produced such phenomena.

Morris (9) recorded the case of a healthy man who fell 20 feet, and developed traumatic emphysema though no broken rib was found. The next day there was a systolic click heard all over the room. It was loudest when the man was turned on his left side and after the fourth day was heard no more.

The havoc of World War I was not without its small and casual by-product of clinical observations on the sound which was given the prosaic but accurate title of pericardial knock. Several British observations begin with that of Captains Rees and Hughes (10) in the *Lancet* in January 1918 Their note, classic in its brevity and clarity, deserves quotation in full.

"We should like to call attention to loud 'tapping' sounds which we have heard over the cardiac area in nine cases of wounds penetrating the left chest, where the missile had lodged near the heart, or where the track of the missile has passed in its close proximity These sounds were quite distinctive and similar in each case, only varying in intensity. They were most marked when the patient was first examined after wounding and persisted for a variable time, occasionally for only 24 hours, but in one patient they were still present on evacuation a fortnight later The sound continued even when the breath was held, although the intensity varied with respiration, increasing during inspiration and diminishing during expiration in all cases It was synchronous with the heart beats, and when loud rendered the heart sounds inaudible *In four patients the sound could be heard with the ear held some inches away*

extraordinary noises "

Colonel S. Maynard Smith's thoughtful commentary on "Pericardial Knock" is reproduced in extenso (11).

"There is a sound heard on auscultation in cases of penetrating wounds of the chest near the cardiac region which is unlike anything met with in civil practice Its

noise which may be compared to that heard in the earpiece of a telephone when the lever is moved up and down It may be heard sometimes when standing at the foot

of the patient's bed. Sometimes the patient is conscious of it, and on one occasion has himself called my attention to it. In nearly every case the sound disappears after forty-eight hours, although in one patient who was recovering from a chest wound without any complications the sound persisted for ten days. He was then sent to the base, and I have no further information about him The sound has been, as a rule, most marked near the apex beat. It is usually double, corresponding to the heart sounds, and its intensity waxes and wanes with respiration, sometimes disappearing completely in full expiration A distinct fremitus can be felt by the hand laid on the chest

"The first case in which I noted it was shown to me by Captain W. W. Rees, R A M C I then suggested that it was due to a shell fragment lodged where the pleura and pericardium are in contact. I thought this possible as I had previously heard a metallic rumbling sound in a case in which a shrapnel ball was seen by x-ray screen in the costo-diaphragmatic angle moving up and down with respiration. A skiagram in Captain Rees's case showed the fragment to be in the neighbourhood of the pericardium

"That this explanation was not the correct one was proved by the case of an officer shown to me by Lieut.-Colonel Newlands of the A.A.M C. *The characteristic sound could be heard when standing beside the patient's bed.* The entry wound was outside the heart on the left side, and the track passed in the direction of the pericardium Two days later a right-sided subphrenic abscess developed and was drained The foreign body was found lying in the right side of the abscess cavity An infected haemothorax was also drained The patient died a few hours later, and at the autopsy a 'shaggy' purulent pericarditis was present This case showed clearly that the foreign body was not itself directly responsible for the sound

"In the next case I saw the foreign body was visible on the screen in the cardiac area It moved continuously with the heart beat, so that a skiagram showed only a blurred image This patient died, and at the autopsy the pericardial surface was normal There was no infection, exudate, or any sign of pericarditis The cavity contained a few drops of blood-tinged serum due to the passage of the missile, which had traversed the pericardium and lodged in the interventricular septum This case, as also several others in which the 'knock' disappeared after twenty-four or forty-eight hours in patients who never had serious symptoms shows that pericarditis is not the cause

"I may note in passing that the continuous movement of the foreign body shadow with the heart beat, as distinguished from an oscillatory movement at certain phases of the respiratory and cardiac rhythm, has indicated that the foreign body is actually in the heart wall or cavity

"It has been [illegible]

over, a case has been reported by Captain Rees in which the sound was heard over the apex beat, which was normal in position It was heard in systole and was loudest in full inspiration The foreign body was seen on the x-ray screen moving with respiration, and in full inspiration there was an oscillatory movement communicated to it by the cardiac impulse There was a clear space of some 2 in between the heart shadow and that of the foreign body The entry wound was over the lower end of the left scapula Localization of the foreign body on anatomical cross-sections showed it to be lying behind the root of the left lung in close proximity to the rami bronchiales, and over 1 in from the nearest point on the pericardium The man's condition when he was sent to the base was very good

"The usual disappearance of the sound after a short period, and the absence of serious symptoms in many cases, suggests that it may be due to air in the interstitial connective tissue of the lung which is struck upon by the beats of the heart. Further records will doubtless throw more light on the subject."

Henley Munden (12) closed out this flurry, bringing up the chance of sport, and the risks of rabbiting.

"A few days before the appearance of Colonel S. Maynard Smith's article in the *Journal* of January 19th, p 78, I was called to see a boy who had been accidentally shot in the head and left side of the chest when rabbiting

"About an hour before he died a loud cardiac 'click' developed which was synchronous with the heart systole and could be heard distinctly six or eight feet away. It bore no relation to the respiratory movements, as the respiration was Cheyne-Stokes in character and the sound persisted during the period of apnoea The simile of the click in the ear-piece of a telephone when the lever is raised illustrates it very well During the last fifteen minutes of life the heart became very feeble and irregular and the 'click' disappeared I was very interested to read Colonel Maynard Smith's explanation of the phenomenon, as I was quite unable to account for it. A postmortem examination was not ordered by the coroner, so no further investigation could be made "

Such phenomena were well known to American physicians in France in World War I as I learned from Dr. Blankenhorn in Cincinnati (13). Apparently no one wrote about it

In 1920 Specht (14) described a case where an injury to the chest produced a disturbance which gave rise to a noise heard clearly one meter from the chest Perhaps Bigger's case belongs here but it is included in the section on pneumopericardium. An old case of Schilling's was reported by Warburg in 1938 (15). A musketeer at some unrecorded post of duty or sport fell from a second story window and hurt his chest Tympany was elicited over his precordium and the *bruit de moulin* was heard.

Funk (1945) has related an unpublished observation on a young Negro man stabbed in the chest with resulting hemopneumothorax "The sounds were clear, loud, snapping, synchronous with systole and audible at the foot of the bed " They lasted for a few hours and vanished (16)

SPONTANEOUS AND INDUCED PNEUMOTHORAX

I cannot find with any certainty the earliest recorded case of sounds produced by pneumothorax Perhaps the first is that of Cornils (1885) who reported loud noises produced by the heart in a 23-year-old man with pulmonary tuberculosis and left pneumothorax (17) They were heard several times by people some distance from the patient Autopsy revealed left- [illegible] There is little doubt that

wheel noise of an ear-

embolism misled some

German physicians into supposing that pneumothorax somehow caused air embolism and thus produced the loud and distantly heard sounds.

Albert (18) reported sounds heard away from the chest in a 24-year-old patient who had been given a refill for a therapeutic pneumothorax. Apparently he thought air embolism was responsible. Boyé, in discussing his paper, reported a similar experience in a patient with a bronchopleural fistula.

This ominous Mühlengeraüsch was reported by Junker (20) in a 26-year-old woman who had spontaneous pneumothorax in association with pulmonary tuberculosis In this same year Lister (21) gave a concise case report, neither embellished nor blemished by speculation It follows.

"*History:* A commercial traveller, aged 31, was first seen on Feb 1, 1928. He complained of slight pain in the region of the second left intercostal space and of a loud tapping sound in his chest He stated that six days previously, while sitting in a tramcar he had experienced a sudden, sharp pain in the left upper part of his chest and had felt faint and breathless for a moment He got out of the tram, and after obtaining a little brandy from a friend who lived near by, he returned home and went to bed as the pain persisted, though he felt quite well in other respects For the next two days he remained in bed and the pain rapidly passed off except for a slight tenderness On the third day he decided to get up but on putting his feet to the ground he was horrified to hear a loud rapping sound in his chest which was also plainly audible to his relatives Accordingly, he returned to bed and the sound ceased when he lay down Next day, he made another attempt, and although the sound returned as before as soon as he stood up, it caused him no inconvenience and he remained up and about and was able to walk around for the consultation two days later without experiencing any ill-effects whatever

"Previously he had been healthy, though he recognized that he was a nervous man At no time had he suffered from pleurisy or cough, nor so far as he knew had he lost weight There was no history of tuberculosis in the family

"*Physical signs* On examination the patient was thin, pale, and nervous, with a spasmodic tic of the right side of the face Pulse 116 per minute, regular. Temperature normal As he sat giving the history of his illness at a distance of some 5 feet a hollow knocking sound could easily be heard coming from the patient's chest Its point of maximum intensity was found to be situated over the fourth left costal cartilage, and a sharp tap corresponding to the sound was felt on palpation over the sternum In character the sound was very similar to that heard over the brachial artery when taking a blood pressure at the point when the diastolic pressure is approached, but was, of course, very much louder It was mid systolic in time, coming at the end of the first sound and separated from the second sound by a short but distinct interval It varied both with posture and with respiration In the prone position it disappeared entirely, on sitting up it became clearly audible towards the end of expiration, but grew fainter and finally ceased as the lungs expanded When the patient stood up the sound was at its loudest, rising to a maximum at the end of expiration and diminishing with inspiration A deep inspiration could stop it entirely, even in the upright position, but ordinary quiet breathing did not do so In other respects the heart sounds were perfectly normal, and the blood pressure was 145 systolic and 90 diastolic.

"There was slight tenderness on pressure over the second left interspace in the

midclavicular line, and the left side of the chest presented the usual signs of a pneumothorax with slight displacement of the heart to the right, so that the right margin of cardiac dullness was [illegible] only 3 inches from the [illegible] splash was heard, nor [illegible] was audible all over the chest.

"A radiographic examination on the fluorescent screen showed the lung to be separated from the chest wall by 1 inch in the lateral plane, and there was also air surrounding the apex and base and lying between the upper half of the lung and mediastinum as far down as the reflexion of the pleura at the hilum The whole lung, therefore, in a partially collapsed condition hung loose in the thorax suspended from the hilum It was kept in constant flicking movement by the beating of the heart so that its free lower margin continually agitated the surface of a small quantity of fluid that lay in the costo-phrenic angle Owing to this constant movement it was not possible to obtain a sharply defined radiogram of the outline of the lung "

I found another case that was published by Wolferth and Wood (22) in 1930 under the unlikely title of *Angina Pectoris*. In discussing the problem of pain in the chest they described a young University of Pennsylvania student who, in the throes of preparing for examinations, experienced pain over the heart. It was paroxysmal, and worse upon exercise It was supposed that a wrestling bout a week previously might have precipitated the small left pneumothorax found on x-ray. On running he had noticed "a sensation as though his heart were 'flopping around loose' inside of him; and five days before, his roommate, while across the room, had heard his heart beating." The heart sounds, clear in the erect position, were not heard when he assumed the recumbent position.

Scadding and Wood (1939) recorded a number of cases (23). The first, a 26-year-old man, had a twinge behind the left shoulder blade. "Four days later, on first waking in the morning, he noted a 'curious bumping sensation' in the chest, accompanied by a faint sound " This was verified by the authors who found a slight left pneumothorax by x-ray The sounds were heard only in the recumbent position. The second, a lad of 17 years, "complained of a heart beat on adopting a 'hunched up' posture; the noise could be heard by his mother a yard away " The sound could be heard off and on [illegible]

[illegible] the region of the apex beat " It vanished when 150 cc more were injected. On the next day, however, she had occasionally noted a clicking sensation in the precordium. The fourth case was that of a 19-year-old youth who had left sided pneumothorax induced for pulmonary tuberculosis and the same click was heard after 150 cc of air were introduced The next day when the patient was placed on his right side "the click became so loud

that it could be heard clearly several yards from the patient without the use of a stethoscope" Twelve days later the sound could be heard "on sitting him up at an angle of 40° and inclining him slightly to the left." Their last case, a retrospective and presumably autobiographical one, was that of a young man in perfect health who had "a sharp pain in the chest, and this was followed by an extremely loud peculiar noise accompanying the heart beat. The noise was so loud that it could be heard in the room below." Three weeks later it disappeared without complication in spite of the diagnosis of rupture of a heart valve. This case, even allowing for the recollection of 20 years, holds the all-time record for loudness as far as I have been able to ascertain. Sharpey-Schafer (24) was moved by this account to put on record the disturbing episode of a 20-year-old man in whom exertion produced pain in the chest going down the left arm Left pneumothorax was discovered and four days later "a loud clicking sound could be heard at the apex synchronous with the heart beat. It was best heard with the patient sitting upright or lying on the left side. It occurred late in systole and was so loud that it was detectable with the unaided ear a foot from the chest." In a few casual notes of a discussion of this problem, there was general agreement that adhesions had nothing to do with the sounds (25).

Three more cases of pericardial knock in pneumothorax were reported by Edwards and Simpson in 1939 (26). A 25-year-old woman had bilateral pneumothorax induced because of tuberculosis and a month after the left side had been injected she observed a pericardial knock. It was heard best when she was lying on her left side. "The sound was audible several feet away, and was synchronous with systole of the heart. It could be stopped immediately by adopting any other posture than the left lateral, or by pressing both hands firmly over the precordial area." Another case was that of an 18-year-old girl with the same sign developing several months after bilateral pneumothorax was induced and adhesions divided Pericardial knock "was not always present . . and sometimes only perceptible on auscultation and was initiated by the patient lying on the left side It was more frequently observed just before a left-sided refill and disappeared when the latter was given." In the third patient, a young woman of 20, "after the induction of left-sided pneumothorax the patient began to experience sensations of a regular 'ticking,' and sounds were audible at a distance of 1–2 feet" Change in posture, pressure and the advent of fluid did not affect the sensations.

Frost and Bing (34) added two cases—the first that of a 17-year-old youth who developed spontaneous pneumothorax on the left while bicycling. There was pain and breathlessness "When the patient was placed on his left side there was a distinct extra-sound which was now and again so

loud that it was audible to the patient and even to other persons one meter away from him. The sound most resembled the clicking of the tongue used to urge on a horse." The other patient was a 19-year-old girl who had a left-sided pneumothorax induced for pulmonary tuberculosis. After pneumolysis "when the patient is lying on the left side series of extra-sounds are now and again audible, sometimes so loud that they may be heard about 1 meter from the patient. The extra-sounds are not heard when the patient is lying on her back."

Interstitial Emphysema of Lung and Mediastinum

It is appropriate that Laennec (5), whose invention of the stethoscope opened the modern era of clinical observation of diseases of the chest, was the first to describe sounds heard at a distance in interstitial emphysema of the lung. Indeed he gave one of the earliest satisfactory descriptions of the lesion itself noting "beads of bubbles along the pulmonary strands." He remarked that childbirth, weight lifting and straining at stool were likely causes. In addition to the dry crepitant rhonchus with large bubbles and palpable crepitation sometimes evoked by pressing over the intercostal spaces, he observed subcutaneous emphysema. He said, "The patients are sometimes sensible of a kind of crackling in the part affected." This statement is the earliest record of sounds audible at a distance. Though Skoda confirmed these observations and Müller emphasized them, neither described sounds heard at a distance.

Petersen (27) recorded a probable case of mediastinal emphysema in a young man of 22 who had pain in the left side after slight exertion. The next day there was a "splashing" sound audible at a distance while he was lying on his left side. It lasted two weeks He quoted Edelfsen (28) as source for another, a young physician who similarly had pain in the chest and a clicking sound heard at a distance. The prompt recovery without sequel suggests that these were cases of pulmonary interstitial emphysema.

There are undoubtedly other early cases on record but the contemporary scene with its numerous papers had its curtain raising in Hamman's rediscovery of the clinical characteristics of interstitial emphysema of the lung (29, 30, 31), the first description of apparently spontaneous episodes and so vivid a discussion of the subject that it at once caught the fancy of physicians That his early cases masqueraded as myocardial infarction emphasizes how severe the accompanying pain may be The story of how he came to unravel the diagnostic mystery, charmingly told by a superb clinician, should be read in the several papers Hamman wrote on the subject. Parenthetically I should mention my fortune in having seen one of his patients when an intern at Johns Hopkins Hospital Excerpts from his papers include the following cases where noises were heard at a distance from the chest:

A 51-year-old physician had pain in the chest while shaving. On the evening of the next day while "lying upon his left side, he heard a curious loud, bubbling sound with each contraction of the heart. His wife sitting upon the bed beside him could hear it very plainly."

A young physician, aged 25 years, had a sudden pain in the left side of the chest and a peculiar crackling sensation in the region of the heart. A few hours later "as he was leaning upon his desk he suddenly heard a curious crackling noise, apparently coming from that part of the chest where the pain was located. . . . It could be heard when the bare ear was held a foot from the chest wall."

A young man of 34 had a pain and choking sensation when getting out of a car. Five hours later he had a fluttering sensation and "heard a peculiar noise coming from the region of the heart and when he put his hand over the heart he felt a peculiar vibration."

It did not take long for publication of subsequent reports of similar cases. Morey and Sosman (1939) told of a husky medical student who suddenly developed a severe pain in the left upper precordial region while working in the laboratory (32) The pain radiated down his left arm and was different from pain associated with pneumothorax which he had experienced previously. On the morning after the precordial pain began, he became conscious of a knocking and clicking in his chest. At times he could feel it and at other times he could hear it "Anyone standing directly in front of him could hear the noise." Somewhat later this year McGuire and I reported two instances of spontaneous interstitial emphysema of the lung and described the patient already mentioned in the introduction (1). The first was a 17-year-old school-girl, who felt a pain in the chest while dribbling a basketball, went home and went to bed "Shortly afterwards she heard a curious noise in her chest and told her mother that she thought she had pleurisy." In the other patient sounds were not heard at a distance.

Wolff (1940) told of a 27-year-old laborer who complained of a peculiar noise in his chest (33). About two weeks previously while rolling an empty wheelbarrow he had suddenly been seized with a sharp pain in the left side of the chest over the heart He reached home an hour later and had intense pain and difficulty breathing "On getting into bed and turning on his left side he experienced a grinding crunching sensation in the region of his heart Standing at least a yard from the patient, his wife heard what she described as a noise like the wadding up of paper." This sound was heard by several other members of the family. When examined, the sound was systolic in time and clearly heard with the bare ear a foot from the patient A small area of pneumothorax was demonstrated with 10% collapse of the left lung

Frost and Bing (1940) reported the case of a 22-year-old woman who developed a painful seizure in the chest (34) She had a sensation of move-

ment in the chest and "when the patient felt this, sounds came from her precordium so strong that they were audible 3 meters away." X-rays and other studies were negative. While the nature of this case is not clear, it is presumably one of interstitial emphysema.

Styron (1941) described a 33-year-old man who while walking was seized with a midscapular pain which radiated around to the precordium, up the left shoulder and finally down the left arm to the wrist (35). There was difficulty in breathing. He was brought to a hospital after three hours. He recalled a similar pain twelve years previously. On his third hospital day the patient complained of a to-and-fro thumping noise. The sound was easily heard at a distance of several feet from the chest wall. When a stethoscope was placed on the precordium, very loud crunching bubbling sounds were heard. The sounds altered markedly with the change in position and were best heard at the left side. The same phenomenal sounds were heard at a later time. In the course of a few hours the sounds disappeared permanently.

Pinckney (36) in 1941 reported the remarkable case of a 24-year-old white woman who had six different attacks of spontaneous pneumothorax, in some of which there was interstitial emphysema. On her third attack she had dull pain and dyspnea "She also noted a loud, clicking, snapping noise audible at all times no matter what position she assumed. In addition when lying on the left side she heard another sound, a crunching, crackling sound like small chicken bones being crushed This sound she cannot say with certainty was synchronous with her heart beat. Both sounds were easily audible to persons in the room with her." On her fourth attack three months later, she developed pain while walking Several hours afterwards there was a loud clicking noise and crunching sound became audible There was a small left pneumothorax by x-ray With her fifth attack 15 months later, she had only a heavy sensation "Again the very loud clicking noise, synchronous with each heart beat, was easily audible to anyone within 20 or more feet, and slightly less audible was the crunching noise when she sat leaning forward on her left side " This lasted for 6 weeks during which time her lung re-expanded Pinckney reported another case, that of a 24-year-old man who developed severe pain while at rest While lying down at bedtime on the third night after the onset of the pain, both the patient and his wife were startled and terrified to hear a loud clicking noise emanating from the region of the patient's heart Upon approaching the patient's bed, this sound became audible at a distance of several feet It lasted only for 24 hours and gradually subsided The presumptive diagnosis was mediastinal emphysema.

Caldwell (37) reported the case of a lawyer who developed pain in the chest 5 hours after a severe paroxysm of coughing. "On awaking he turned

on his left side and became conscious of a noisy thumping in his heart. He was able to work . . . although this painful thumping and noise, which even his friends were easily able to hear, persisted." It could be heard many feet away and it lasted one week.

Miller (38) reported a 27-year-old resident physician at the Cincinnati General Hospital who developed a sudden knife-like pain in the chest while making ward rounds. He was put to bed. A small left-sided pneumothorax was discovered. Two days after the painful attack, he had recurrence of sharp pains over the left precordium. "Soon after this he heard peculiar loud clicking noises in his chest synchronous with the heart beat. Palpation by the patient over the sternum revealed a thrill was present. The patient's wife who was standing five or six feet away from the bed was also able to hear these sounds."

Meek, in 1942, reported a 27-year-old white man who developed soreness in his chest after lifting storage batteries (39). "The soreness disappeared in two or three days and a clicking sound, synchronous with the heart beat, was noticed when he was lying on his left side This sound was present in no other position" The physician heard plainly a clicking synchronous with each heart beat that could be heard very plainly and even felt. It began four to six days after the initial strain and lasted several weeks.

Griffin (40, 41), in 1941 and 1942, described two patients, the first a 30-year-old white man who came in to have his heart examined because two months previously he had become conscious of a snapping sound and a vague discomfort in his chest. "The sound was audible to him and to others." He stayed in bed for two weeks The diagnosis was pericardial adhesions. The sound recurred and the pain in the right shoulder returned. There was a slight amount of air over the left apex by x-ray The sounds were audible without the stethoscope when he was in the recumbent position. The second patient was in 18-year-old college student who experienced a sharp pain near his heart while sitting quietly studying. Four days later he was quite uncomfortable and he became aware of something shifting in his chest with change of position "He became conscious of a crunching sound in his chest."

In 1943 several reviews appeared, first by Lintz (42) who quoted many of the cases published up to that time. His case report was that of a 22-year-old automobile mechanic admitted to the hospital one hour after the onset of left-sided chest pain He had had some difficulty in breathing When he was examined he stated that he heard noises in his chest which sounded as if gears were grinding together. "Boiler-like" sounds of loud intensity were heard. They might be compared "to the noise that one hears on crumpling a handful of cellophane close to the ear and could be heard distinctly several feet from the patient's chest." No correlation was noted between the heart

beat and the sounds in the chest. The sounds lasted less than 24 hours. Greene (43) reviewed the cases presented previously and reported some very ingenious observations and novel ideas worked out in association with Barnwell on the mechanism of sounds audible at a distance from the chest Clicking and tapping sounds heard in mediastinal emphysema were thought to be due to the heart's impingement on large emphysematous blebs. He reported a case of a 41-year-old Negro physician who while sitting at his desk developed substernal pain which was severe and radiated to the left shoulder. He went to bed immediately and noted that lying on the left side produced more pain and caused tapping, crunching, and bubbling sounds to come from the chest. These sounds were audible to his wife at the bedside and were synchronous with the heart beat. Later an x-ray of the chest revealed air in the anterior mediastinum.

In 1944 Miller (44) reported spontaneous mediastinal emphysema in a 23-year-old soldier who "heard peculiar sounds in his left chest."

McCabe (1947) discussed the problem of the differential diagnosis with emphasis on the simulation of organic heart disease (45) In the addendum to his paper he reports 4 patients who had interstitial emphysema, 3 with a left pneumothorax in whom the presenting complaint was "a sharp pain, sudden in onset, and felt usually in the left shoulder area, moderate dyspnea, tachycardia and a peculiar crackling sound over the precordium." Dickey's report of spontaneous mediastinal emphysema and pneumothorax in a student health department in 1948, records a number of instances where sounds were heard by the patient or by neighbors (46) One was a 19-year-old student With the second of two episodes of spontaneous mediastinal emphysema, he was aware of noise in the substernal area, although it is not clear that it was heard by others Case 2, a 21-year-old man had severe precordial and retrosternal pain at the level of the 4th rib, which began while he was studying He had some shortness of breath and "he had also been conscious of a grating over the precordium on several occasions. . . . Loud precordial knocking sounds were audible at times without the aid of a stethoscope " Case 3, a 26-year-old woman student had pain in the left chest and substernal region radiating into the neck and left arm "She was aware of a sensation of rumbling in her chest." Case 4, a 26-year-old student had a sudden pain in the left anterior chest increased by deep breathing and accompanied at times by a sound "like tissue paper crackling " A small left pneumothorax and mediastinal emphysema were demonstrated by x-ray.

Although he did not specifically comment on noises heard at a distance from the chest, Macklin's extensive observations on the mechanism and significance of mediastinal and interstitial emphysema of the lung have clarified many of the vexing clinical problems in this disorder (47) In

particular emphasis has been put on the occasional serious case where an air lock with pressure and distention of the interstitial spaces impedes circulation through the lung to a serious degree. Unless it is treated by some form of decompression plus oxygen therapy it may lead to death

Aisner and Franco (48) in 1949 reported sounds heard at a distance from the chest in an 18-year-old college student, an athlete, who developed a sudden, sharp, stabbing, knife-like pain over the precordium while walking. The pain became progressively more severe, was aggravated by breathing, was not relieved by rest and "was associated with what the patient termed a scraping noise in the chest".

During the pandemic of influenza in the latter part of World War I, many observers reported interstitial emphysema of the lung and pneumothorax complicating influenza and its attendant pneumonary sequels. Although subcutaneous emphysema and pneumothorax were recognized, apparently no one described Hamman's sign and apparently no noises were heard at a distance from the chest. At least none was recorded in the reports of Torrey and Grosk (49), Clark and Synnott (50), Berkely and Coffen (51), and Bullowa (52).

Mediastinal emphysema and pneumothorax complicating parturition has been recorded as a not too rare complication In the review of Gordon (53) in 1927, and that of MacRae (54) in 1949, no mention is made of sounds heard at a distance from the chest although the characteristic noises heard by auscultation were described in many instances.

Traumatic Mediastinal Emphysema

It is not always clear from descriptions whether certain traumatic cases in which precordial noises were heard at a distance from the chest should be put in the category of mediastinal emphysema or pneumothorax Steiger (55), in 1864, described a man who fell, fractured his arms, shoulders, and ribs He had marked precordial tympany and a splashing, metallic sound synchronous with the heart beat was heard at a distance from the chest Since this patient recovered it is not known whether he had pneumothorax, pneumomediastinum or mediastinal emphysema but in all probability he represents the same type of case where pericardial knock is heard In 1875, Leonspacher (56) described precordial tympany and musical heart sounds heard 3 feet from the bed in a man who had fallen on his back He too recovered and the exact nature of his lesion was not known. Weil (57) recorded an instance of mediastinal emphysema in a young man who was given an injection of novocain into the brachial plexus, and sounds were heard at a distance for a few minutes. Jehn (58), in 1921, reported an 18-year-old man who fractured his larynx Sounds synchronous with systole and diastole were heard at a distance from the chest. After death air as well as pus was

found in the mediastinum. Hörnicke (59) described a 33-year-old man who was injured, developed a subcutaneous and mediastinal emphysema. The sounds were heard at a distance. This case was verified at autopsy. Bigger (60), in 1932, described a 22-year-old Negro who had a gunshot wound of the precordium. Slight subcutaneous emphysema was found and "six hours after admission a peculiar splashing sound was discovered over the precordium synchronous with the heart beat." X-ray showed traumatic pneumopericardium

ALIMENTARY CANAL

Rupture of the Esophagus

In 1722, Boerhaave (61) reported the first case of rupture of the esophagus. "We have the surprising observation given us by the celebrated Boerhaave, which is perhaps the only one published, namely the illustrious Baron Wassenaer, Lord High Admiral to the Republic, after intense straining and vomiting broke asunder the tube of the esophagus near the diaphragm so that after the most excruciating pains the aliments which he swallowed passed, together with the air, into the cavity of the thorax, and he expired in 24 hours" Unfortunately, there is no comment upon any sounds heard at a distance from the chest In a number of recent reviews which have emphasized the importance of diagnosing rupture of the esophagus because of the possibility of surgical relief of the condition, there is no comment on noises heard at a distance from the chest. Insofar as I have been able to find, the only clearly demonstrated case was that of Begbie (62), published in 1863, in which he described a patient with carcinoma of the esophagus which ruptured into the pericardium, and splashing and churning sounds were heard at a distance from the chest In all likelihood this should be included under the heading of pneumopericardium rather than here However, since diagnosis of rupture of the esophagus is urgent the possibility that it may produce sounds heard at a distance from the chest should be borne in mind in the differential diagnosis Interstitial emphysema or pneumopericardium may actually cause loud sounds under such circumstances

Ectopic Borborygmi

Portions of the esophagus, stomach and colon may contain trapped air which produces sounds heard at a distance where the heart beat provides the percussion force. At times one can count the pulse from such sounds, though there are apt to be variations produced by respiration, change in position and belching or expulsion of flatus

A unique case of precordial noises reported in a brief note published by Allen (63) is quoted in full. "A woman came for examination complaining

of 'heart trouble' of unusual nature. She had been disturbed by a noise synchronous with the heart beat which had appeared intermittently for two years. It frequently bothered her during the day but was likely to be particularly troublesome when she was lying down at night. The sound was so loud that it was heard for a considerable distance from the body. Not only the patient but other people could hear it; the latter even before the patient entered the room. She described it as resembling the splashing of water in a mill. The cause of the noise was discovered more or less by accident. She had moderately severe anemia which had not responded to treatment. X-ray examination of the gastro-intestinal tract was made in order to see if there was any lesion responsible for loss of blood. It was found that she had a diaphragmatic hernia; the large part of the stomach was lying in the chest. The sound was produced through action of the beating heart on the contents of the intrathoracic stomach."

Ryle (64), in his book, *The Natural History of Disease*, refers to two interesting cases, the nature of which is not entirely clear.

"A well-built nervous man, age 49, with a family history of gout in his father and asthma in his son, the personal history of gout, migraine and iritis, complained that for 10 years he had been liable at long intervals to attacks of severe pain at the lower end of the sternum radiating up into the chest and jaws He declared that it would be unendurable if it lasted more than a few seconds This description at once suggested angina pectoris but further inquiry showed that it had never been induced by walking or effort, that it generally came on while he was stooping over his desk, that it could be immediately relieved if he was able to swallow some fluid or solid, and that on swallowing he heard *a distinct click* as the pain departed With reassurance and gentle treatment the symptoms remained in abeyance for many months The patient later consulted me for extrasystoles "

"A nervous Welsh woman giving a family history of insomnia from which she herself suffered and of 'nervous breakdown' in a sister and shellshock in a brother complained of a sensation of 'a lump in the throat ' This would be worse at night and on sitting up she maintained that she could hear food *'trickling through it'* On these occasions she had brought up food She had always experienced difficulty in holding her water X-ray examination with emulsions of varying consistency failed to reveal any abnormality of the gullet "

Greene (43), who has contributed notably to the study of such noises, describes some curious observations In one patient with a left pneumothorax who was being prepared for introduction of more air a metallic knocking sound began to emanate from the patient's chest audible at a distance of 3 feet Several minutes later borborygmus was heard and the sound disappeared without introducing more air or changing position. In this and another patient introduction of air into the colon, sufficient to distend the splenic flexure, again produced the noise which was of a metallic knocking quality, sometimes heard in systole, sometimes in diastole. It was concluded that it was caused by rotation of the heart which struck the

diaphragm immediately above a bubble of gas in the colon if systolic; and caused by the free fling of the uncushioned heart in diastole striking the diaphragm if diastolic.

Roberts (65) describes some observations which perhaps should be included in the final section on bizarre and miscellaneous noises. "A *gastro-cardiac splashing sound* may be produced by (1) a large overactive heart with (2) a suitable mixture of gas and fluid in the stomach. We have heard similar transient *'cardio-esophageal' sounds*, at times quite alarming, in three patients who had air and fluid in the esophagus as demonstrated by fluoroscopy. Apparently a combination of transient cardiospasm, drinking of liquid and air-swallowing had trapped the air in the esophagus so that it bubbled with each heart beat The sound (bubbling, loud, and at times audible to the patient) was heard best over the midsternum, but shifted its level of greatest intensity from moment to moment Similar phenomena may occur over an esophageal or gastric hiatal hernia."

HEART AND PERICARDIUM

Pneumopericardium and Hydropneumopericardium

The confusion concerning the significance of heart sounds was just beginning to lessen when Laennec (5) wrote the following paragraphs on "pneumopericardium":

"By this expression I shall designate those collections of air, howsoever produced, which are met with in the pericardium They are very often observed in the examination of dead bodies, particularly such as have been kept some time In the latter case, the effusion is, no doubt, the effect of decomposition, but in many others the complete absence of all signs of putrescence proves it to have existed previously to death. Sometimes the air is combined with a liquid, and this is by much the most frequent case, at other times, the pericardium is distended by air alone The effusion of air and serum into the pericardium, may occur in the agony of all diseases I have sometimes been enabled to announce its presence, from the supervention of an increased resonance over the lower part of the sternum, and from the existence of the sound of fluctuation produced by the action of the heart, and by deep inspirations

As these observations were anterior to those made respecting the sound of the heart's action heard at a distance from the body, I did not ascertain whether this last-mentioned phenomenon was present or not but I am convinced that in almost [illegible]

[illegible] conceive only four capable of giving rise to it 1) that just mentioned, 2) the devel- [illegible] roposition altogether inad- [illegible] such a state, 3) the ossifi- [illegible] the sternum or cartilages of the ribs—a condition of parts incomparably more rare than the phenomenon in

question; lastly, 4) the co-existence of such a degree of induration of the muscular substance of the heart with such violent action of it, as to render its impulse against the thoracic parietes (that is, the contact of two surfaces comparatively soft and moist) productive of a sufficient degree of resonance. This last hypothesis becomes the more improbable from this consideration, that when the heart is indurated it is also hypertrophied; and we know that the persons in whom the sound of the heart is heard at a distance, are almost always nervous subjects, with a soft muscular fibre, and a heart possessing very little real force of contraction."

Even before this time Morgagni (66) spoke of having heard the splash of water and air in the pericardium and Portal (67) had encountered this combination at autopsy without recording clinical findings.

The first real advance in this topic was the observation of Brichteau (1844), who described a Polish veteran of Napoleon's army who was struck down by a blow on the chest by a carriage tongue (68) His wife heard "boiling" in his chest and Brichteau and 8 assistants all heard it. The man died and autopsy disclosed pericarditis with the profuse formation of evil-smelling gas.

Stokes' (1855) observations speak for themselves (69):

"There seems no reason to believe, that if air be occasionally produced in the pleura or peritoneum, when in a state of irritation, that the same should not occur in the pericardium On this subject I have no anatomical evidence to produce, but I feel satisfied that in one case at least I observed the phenomena of pericarditis with pneumatosis The patient was a young man of lymphatic temperament, who had laboured under an attack of acute pericarditis for a few days before I saw him On my first examination he presented the usual signs of dry pericarditis, with a considerable effusion of lymph of the ordinary consistence The rubbing sounds, though loud and distinct, had nothing unusual in their character, and the patient suffered but little distress After two or three days I saw him again, and found that his state had become very much altered His appearance was haggard and worn, and he complained of extreme exhaustion, which he attributed to a total deprivation of sleep This was induced by the extraordinary loudness and singular character of the sounds proceeding from the cardiac region, for though up to this period the rubbing sounds were distinctly perceptible by means of the stethoscope, the patient was quite unconscious of their existence They had suddenly, however, become so loud and singular, that the patient and his wife, who occupied the same apartment, were unable to obtain a moment's repose On examination, a series of sounds was observable which I had never before met with It is difficult or impossible to convey in words any idea of the extraordinary phenomena then presented They were not the rasping sounds of indurated lymph, or the leather creak of Collin, nor those proceeding from pericarditis with valvular murmur, but a mixture of the various attrition murmurs with a large crepitating and a gurgling sound, while to all these phenomena was added a distinct metallic character In the whole of my experience I never met so extraordinary a combination of sounds The stomach was not distended by air, and the lung and pleura were unaffected, but the region of the heart gave a tympanitic *bruit de pot fele* on percussion, and I could form no conclusion but that the pericardium contained air in addition to an effusion of serum and coagulable lymph "

Aran (70) described the same phenomenon in a man whose pericardium was punctured by a trocar.

In 1862, Niemeyer (71) quoted a case reported by Tütel of carcinoma of the esophagus with perforation into the pericardium. "Even at some distance from the patient a peculiar, clear splashing sound can be heard, which comes and goes with short rhythmical intervening pauses, and which beyond all question, is caused by the agitation of the liquid contents of the pericardium by the movement of the heart. In my case this splashing sound was distinctly audible to the roommates of the patient, who lay at the other end of the ward."

The most detailed and clear-cut study of the problem was reported by Morel-Lavallée (7) in 1864. After some shrewd speculation he reports several cases The first was a 20-year-old carpenter previously healthy who fell from the 7th floor, hit a projection on the 4th and crashed to the ground. Shock, fractured left leg, incomplete paraplegia and hematuria ensued. On the next day the man said that during the night he had been wakened several times by a noise coming from the left side of the chest resembling that produced by blowing into a wide bottle It lasted 7 days and was never heard in a sitting position He died suddenly while trying to sit up on his bed. Autopsy showed multiple ruptures of the pericardium with air in it, slight pneumothorax and communication between pericardium and pleura. Two kinds of noise were heard, one the millwheel sound, and one a kind of metallic tinkle. Another was a 45-year-old coachman who had fallen beneath a horse which stepped on his chest. A bruise the size of the hand was found over the sternum Bony crepitus was detected After a stormy course precordial sounds increased in intensity and were heard one meter from the chest Autopsy revealed widespread sepsis, with traumatic pyopneumopericardium.

In 1873 Eisenlohr (72) recorded the case of a 30-year-old woman who was suffering from pyopneumothorax and pneumopericardium. A loud splashing sound was heard at a distance from the chest Friedreich (73) recorded a similar phenomenon in a woman who had pleurisy and empyema following a difficult labor. A metallic sound produced by the heart's action was heard at a distance Precordial tympany was found. In both instances autopsy revealed air in the pericardium along with pus

According to Gibson (70) Walshe recorded the unhappy accident of a sword swallower who pierced his pericardium through the gullet and had a noisy pneumopericardium

Fetzer (74) observed a man whose shoulder had been injured by a bullet wound Suppuration and arthritis supervened Precordial tympany was detected and musical heart sounds were heard at a distance Autopsy disclosed air and pus in the pericardium but there was no perforation of the pericardium.

Meigs (75) reported in detail the findings in an 18-year-old boy who developed pyopneumopericardium after his esophagus was perforated by a small chicken bone. He remarks that "One of the family called my attention to a very peculiar sound in the patient's chest, which was audible to a person sitting by the bedside (three or four feet off). It . . . had a splashing character, and suggested to myself the sounds produced in an old-fashioned upright churn, which, being worked by the arms had a certain regular rhythm. Listening over the heart, both the sounds and the friction murmur were accompanied by the most extraordinary metallic reverberation I had ever heard, and were followed by the loud splashing and churning sounds which had been heard at the bedside. The echo-like reverberation was so like the amphoric note heard in pneumothorax, and the churning sound so like the succussions of the same disease that the presence of air and liquid in the pericardium was at once suggested to my mind."

Guttmann (76) reported a somewhat similar source of air in the pericardium in the case of a 36-year-old laborer who had a penetrating ulcer of the stomach which perforated into the pericardium. There was precordial tympany and the loud metallic heart sounds were heard at a distance from the chest. There is a review of many of the previously reported cases in his article.

Love (77) described a 28-year-old bank clerk who experienced pain below his left nipple, cough, and fever of 100°F. A pericardial friction rub was heard. On the fifth day "the patient's wife called attention to a loud click which she heard, and I then heard at a distance of two or three feet from the patient. It was ventricular systolic in rhythm." Eleven days later it could be heard 20 feet away. It suddenly vanished 19 days after it came and was superseded by a friction rub

Nicholls (78) recorded the occurrence of pneumopericardium as one of the complications in a 21-year-old man who had gas bacillus infection with peritonitis after a rupture of the appendix. Nine days after the operation crackling was felt over the precordium which bulged and on the next day loud crackles resembling the metallic tinkle in pneumothorax were heard synchronous with the heart beat. "Splashing sounds could be heard some distance from the patient." They lasted only one day. Two days later he died and at autopsy gas and foul-smelling fluid were found in the pericardium.

Laub (79) put on record a somewhat similar finding in a 22-year-old man whose infection began with facial erysipelas Precordial tympany was detected with shifting dullness below and a low metallic splashing sound was audible at a distance.

Another instance of pneumopericardium complicating perforation of stomach ulcer into the pericardium was reported by Saexinger (80). Pre-

cordial tympany gave the first clue to the complication and a metallic splashing murmur was heard at a distance.

James (1904) gave a very extensive survey which included most of the previously reported cases along with a case report of his own (81). A 25-year-old Irish woman developed pneumopericardium as a sequel to injury of the esophagus from a fragment of mutton bone. The pertinent observation: "When the patient takes a full breath and holds it there is heard by the listening ear, held twelve inches from the chest wall, a peculiar clicking sound synchronous with cardiac systole; this click has a musical quality with the quality of a succussion sound or splash. It disappeared with extreme expiration. Upon applying the ear or stethoscope to the precordium the heart sounds themselves are dull and muffled, but there are certain remarkable and unusual phenomena. The first sound is accompanied by a loud metallic tinkle of splashing, gurgling quality, suggesting the sound made by an old-fashioned over-shot waterwheel, the *bruit de la roue hydraulique*. It is as if a very marked succussion were brought out by each systolic contraction of the heart, together with many metallic tinkles ... Its intensity varies from time to time without apparent reason."

Meyer (82) reported "spontaneous" pyopneumopericardium in a 29-year-old white man. Sounds could be heard a foot and a half from the chest but 10 days later were heard only 6 inches away. Later they disappeared. The cause is obscure.

Yates (83) reported the case of a young farmer who fractured his fourth and fifth ribs on the left when thrown from a wagon. About 10 hours later, "during the night there had developed with each cardiac impulse a well-pronounced splashing or succussion sound It was synchronous with the systole, and loudest just above the apex beat This sound was loud enough to be distinctly audible all over the room . The abnormal sound persisted for some ten days or two weeks "

Heise and Brown (84) reported hydropneumopericardium in a 26-year- [illegible]

his left side. Later there was a splashing sound which could be heard 6–8 cm. from the chest. The splashing and heart knock sound persisted for 14 days.

Stahl and Entzian (85) removed some pus from the pericardium of a 51-year-old worker with tuberculous pericarditis and injected air The characteristic waterwheel sound was produced This is the earliest note I have found of injected air producing this sound though probably there are other reports. These workers produced air embolism in dogs and heard sounds 20 inches away.

Another extensive review of the subject of hydropneumopericardium was reported by Shackelford (86) in 1931. His patient was a 61-year-old machinist. Two weeks after a bout of pericarditis "the patient was feeling quite well, got up to shave, and while leaning forward in the act of shaving he heard a metallic, clicking sound in his chest." The noise was audible to others 4–5 feet away and lasted for 10 days

Gilbert (87) reported an instance of a 20-year-old man who had a pain near the heart after lifting a bale of hay. A few minutes later he heard a peculiar noise in the chest. "These sounds are forcefully audible over the entire room." X-rays revealed air in the pericardium but how it got there was a mystery. It is possible that this was really a case of mediastinal emphysema with a large bulla.

Trimble, Eaton and Thompson (88) reported spontaneous pneumopericardium in a 23-year-old man with apparently arrested pulmonary tuberculosis. A day after noticing pain beneath the sternum he had "a gurgling sensation in this area associated with each heart beat." Air in the pericardium was demonstrated by x-ray.

Price (89) described the findings as follows: "On auscultation, metallic and splashing sounds, resembling those produced by a water-wheel or a churn, synchronous with the movement of the heart, are audible; these sometimes become more pronounced on shaking the patient, and occasionally may be heard at a considerable distance "

Air Embolism

It is not surprising that air embolism is a subject about which there is much rumor and general discussion and relatively few case reports, since air embolism indicates an accident In some cases negligence or carelessness is suggested. The effects of injection of air into the veins, arteries, and heart of experimental animals had been known for a long time, and occasional accidents, particularly in connection with operations on the thyroid and the neck, had made surgeons well aware of the alarming and disconcerting sounds which were produced by air embolus. Gundermann did work along these lines. Several reports from German medical authors indicate a possible confusion in the case of sounds associated with pneumothorax, particularly artificially induced pneumothorax. Wagner (90) reported an instance of sounds heard at a distance from the chest in an operation for retrosternal goiter when air was introduced into the left innominate vein and air embolism resulted The cases of Hirsch and Sauser (91) probably represent an instance of pneumothorax without air embolus in the first case and a combination of both in the second case, that of a 42-year-old woman who had a refill for a pneumothorax and who then had sounds heard a meter away from the chest In all probability there was air embolism

because she developed temporary hemiplegia. This point cannot be determined with clarity. Durant, Long, and Oppenheimer (92) discussed this problem in their article on pulmonary air embolism and stated, "In the pulmonary or venous form the presence of air in the right ventricle produces a loud churning sound often readily heard without stethoscopic aid which is known as the millwheel murmur. This murmur appears almost immediately after air has entered the venous circulation." Crile (93), in his book on thyroid disease, states that, "Air embolisms may cause no symptoms or may give rise to an audible churning sound when the air reaches the heart. This persists for about 15 seconds until the air is absorbed" DeGowin, Hardin, and Alsever (94) discussing this problem as a complication of blood transfusion and blood donation remark that, "An observer as well as the patient may hear a gurgling or clicking sound in the heart . . . a millwheel murmur may be heard in the heart." In the majority of articles on this subject there is no mention of the fact that sounds were heard and the individual case reports are very rare. In 1950, Stallworth, Martin, and Postlethwait (95) described a patient who developed air embolism as a consequence of an operation or an infusion of blood during deep shock, and the characteristic sound was described as a millwheel murmur. Although it was not explicitly stated that this was audible at a distance from the chest, a letter from Dr Stallworth mentions that once it was heard with the stethoscope close attention permitted the observers to hear it without the stethoscope

Heart Murmurs

Before the introduction of the stethoscope focussed the clinician's attention on sounds produced by the heart there were references to audible precordial sounds found in conjunction with heart disease Some of these are described so vaguely that they are of little value Others probably merit inclusion in what has now become a formidable list of endocardial murmurs heard at a distance by the unaided ear Allan Burns (96) described a case seen by Dr Brown in the Edinburgh Hospital, a man with dropsy who had "a jarring when the ventricles contracted; and when the hand was laid on the side it resembled the feeling of a varicose aneurysm, his expectoration when he used exercise was bloody, he had unusual palpitation, jarring sensation and hissing noise, as of several currents meeting; the sound was frequently audible as in the varicose aneurysm" Mitral stenosis, pericarditis and induration of liver and lungs were found after death Burns referred to the phenomenon as "audible palpitation", and the presumption is strong that the sound was heard at a distance though not explicitly so stated.

Corvisart (97) in 1818 described the phenomenon but had encountered such noises only once.

Laennec (5) had given this matter detailed study, and his observations are so unusual that one must ascribe his findings to marvelous acuity of hearing or being carried away by rhetorical enthusiasm. The following quotation is from Forbes' translation.

"It had long been believed, but rather on the faith of traditional report than from actual observation, that the pulsations of the heart may be sometimes heard at a certain distance from the patient. Corvisart informs us that he had observed this fact but once, and only then on placing the ear very near the chest. Many years since, I was informed by several patients, that they were subject to palpitations of such severity that they could be heard at the distance of several paces; and one of these patients, as well as persons of credibility, witnesses of the fact, assured me that in his case the palpitations, could be heard in the chamber adjoining that in which he slept. I observed this phenomenon for the first time in the year 1823, in the case of a

case have I myself heard the pulsation at a greater distance than a foot and a half or two feet, but we can readily admit the possibility of this I have several times ascertained from the perfect accordance of the sound with the pulse, that it was owing to the contraction of the ventricles I do not recollect to have ever heard it produced by the auricles Out of more than twenty subjects in whom I have heard the pulsation at a distance of from two inches to two feet, three or four, at most, were affected with organic disease of the heart All the rest labored under palpitation of a purely nervous kind, and several were only so affected after quick walking, or ascending a staircase In all of them the effect was temporary, and several, after a certain time, regained perfect health The bellows-sound and purring thrill frequently exist, in a slight degree, particularly in the arteries, in such cases Never having had an opportunity of examining the body of anyone who had presented this phenomenon, I cannot speak with any certainty as to the organic cause of it, but I am induced to consider it as owing to the presence of a greater or less quantity of air in the pericardium The ossification of some external part of the heart, may also give rise to the phenomenon, but I have met with no example of the kind"

Andral's commentary on this interpretation is worth including (5)

"Laennec's explanation of these sounds of the heart heard at a distance, appears

borygmi developed *in the intestines filled with air and liquids, to the sounds of the* heart? The two facts cannot be connected unless we hear in the heart sounds similar to those caused by the displacement of gas It is yet to be proved also, that the artic-

uals can produce at pleasure These are only frictions between the surfaces of the joints. the same sound is heard in the pericardium when the false membranes rub together on its inner surface."

In the classical description of hyperthyroidism which has given him eponymic fame, Graves (98) encountered the phenomenon of heart sounds heard at a distance.

"I have lately seen three cases of violent and long continued palpitations in females, in each of which the same peculiarity presented itself, viz. enlargement of the thyroid gland; the size of this gland, at all times considerably greater than natural, was subject to remarkable variations in every one of these patients .. The palpitations have in all lasted considerably more than a year, and with such violence as to be at times exceedingly distressing, and yet there seems no certain grounds for concluding that organic disease of the heart exists In one the beating of the heart could be heard during the paroxysm at some distance from the bed, a phenomenon I had never before witnessed, and which strongly excited my attention and curiosity She herself, her friends, and Dr Harvey all testified the frequency of this occurrence, and said that the sound was at times much louder than when I examined the patient, and yet I could distinctly hear the heart beating when my ear was distant at least four feet from her chest! It was the first or dull sound which was thus audible This fact is well worthy of notice, and when duly considered appears to favour the explanation lately given by Magendie of the causes of the sounds produced during the heart's action, for none of those previously proposed seem to be capable of accounting for a sound so loud and so distinct "

In his remarkable book which might be called the philosophy of teaching heart disease, Latham (99) detailed his experience with such exuberant noises Among the factors making for loudness he lists "a peculiar quality of the endocardial murmur, giving it a high musical note Such a murmur will sometimes refuse to suffer restriction to any certain space within the body It will even carry itself outwards and reach the ears of bystanders at a short distance "

Quain (quoted by Howard, 100) described a case where a murmur heard several inches away from the chest occurred following a strain

Peacock (101) described a loud musical diastolic murmur in a 64-year-old man "It exactly resembled the sound produced by the common cuckoo clock" (now alas nearly extinct) and was heard at a distance of several feet The autopsy revealed thickened aortic valves separated from each other The free edge of the right leaflet was retroverted

Stokes (69) reported two instances of murmurs heard at a distance in "extensive and irregular ossification of the aortic orifice " The sounds were audible to the patients, and with one "the perception of these sounds was the principal cause of his suffering for his general health long continued excellent, and the heart's action was but little excited This gentleman once observed to me *that his entire body was one humming-top*. The loudness of

the tone varied with the force of the heart. When I first saw him the sounds were audible at a distance of at least three feet, but when the force of the heart had been reduced . . . the loudness of the sound at the aortic orifice was so much reduced as to render it inaudible, unless by applying the ear." Humming was heard over the limbs, probably transmitted by bone.

Simpson (102) described a 40-year-old man who, while helping to move a piano down a flight of stairs "suddenly felt something give way in his chest, and immediately heard a noise proceeding from it which he compared to that 'made by a young frog'." It was audible to those about him as well as himself. It could be heard for 9 months and then faded out. Four and a half years later he died of heart failure, having signs of free aortic regurgitation. The diagnosis of rupture of an aortic valve was made.

In 1874 Yeo (103) described a 45-year-old laborer who, following injury, complained that he heard a loud "singing" noise within his chest "and indeed it is not difficult for other standing near him to hear the sound which he hears himself." The sound was heard three feet away and signs of aortic regurgitation were found. He referred to Dr. Beale's case of similar nature which followed shovelling. There was sharp pain and "a rhythmical humming noise was clearly heard without putting the ear to the chest, as far off as eight inches from the surface of the body." It was diastolic in time.

Orton (104) described a man of 40 who was engaged in weight lifting when his trouble began "He could hear a sound in his chest which he described as like a pigeon cooing."

Frew and Finlayson (105) related the distressing sequel to a brawl which befell a 39-year-old man Though no blow was struck there was much straining and struggling There was a " 'cooing sound' in his chest resembling the soft cooing of doves . . . This cooing sound was also heard so plainly by his wife that she complained of its disturbing her while in bed with him " It was heard readily 1–2 feet away from the chest, and there were signs of aortic regurgitation The sound could be heard for many months but was gone 19 months after the onset. He died in congestive failure 34 months after the accident

Barie (106) related the misadventure of a 45-year-old worker who was thrown from a carriage, hitting his chest on a rock "A very rough diastolic murmur could be perceived 50 centimeters from the chest wall" and was disturbing to the patient himself. It was diastolic in time, and with other signs pointed to the diagnosis of traumatic rupture of the aortic valve

A similar case was discussed by Tretzel (107) in a 41-year-old laborer. The murmur was heard three meters away

Langwill (108) heard a loud systolic murmur 5 inches from the chest wall in a 19-year-old youth who died in heart failure Subaortic stenosis and scars from old endocarditis were found at autopsy.

Marshall (109) reported a 42-year-old man who fell on his chest and "complained of a loud noise in his chest." He had a very loud diastolic murmur. The diagnosis of rupture of the aortic valve was made.

Allbutt (110) refers to aortic stenosis where the murmur "may be loud enough to be heard at a distance from the chest." He recorded, without details, a case where the murmur was heard one inch away from the chest; and commented that such murmurs were surely aortic.

Hoffmann (111) reported a murmur heard some distance from the chest in a man who had rupture of the aortic valve

Wilson and Jamieson (112) reported three interesting examples of musical diastolic murmurs heard at a distance from the chest in soldiers two of whom had experienced trauma, though the noises were not heard until some time later. "They were all sufficiently loud to be audible to the patients themselves and to the unaided ear of the examiner at a distance of from one to five feet from the chest wall" Case 1, a 45-year-old butcher had been healthy and engaged in strenuous sports. While convalescing five weeks after being buried by an exploding shell, he became aware of a "whizzing" sound in the chest. "In a quiet room the murmur could be heard with the unaided ear at a distance of four feet from the chest." *Case 2, a 42-year-old brewery cellarman turned soldier became conscious* of a persistent noise in his chest "like water rushing through a pipe" four days after being gassed (phosgene) He was aware of it only when in the horizontal position There was much variability in the intensity of the sound. Peripheral signs of free aortic regurgitation were found and the blood pressure was 135/45 Case 3, a 34-year-old former dock laborer was thrown about by the explosion of a large shell but not obviously injured. After a six-month period of nervousness and palpitation during which he was able to do heavy work, he became aware of an intermittent noise in his chest, louder after exertion "It could be heard readily in a quiet room at a distance of 8 inches from the chest" The blood pressure was 140/60. The murmur was louder when he was lying down There was a suspicion of syphilis in two of the men and the traumatic episode in two Nothing is known of the subsequent fate of these patients

In his classic study of rupture of the aortic valve C P Howard (100) described 21 cases where the murmur was heard by the patient and his friends This number represented about a third of the non-traumatic cases The variations in loudness of the sound are illustrated by Quain's case (heard several inches from the chest), O'Neill (6 feet), Dupuis (15–20 centimeters); Tranquilli (50 centimeters), Schneider (25 centimeters) and Schlecht (several centimeters) Along with other examples the murmur was compared to the "croaking of a frog", the "cooing of a dove or pigeon", a "rumbling, rustling noise", a "humming noise", a "whistling noise", a

"buzzing in the chest", a "musical murmur or thrill", a "whining noise" and even a "rattle in the head".

In the 1937 edition of his book on physical diagnosis, Major (113) comments on cardiac murmurs heard away from the chest. "In a patient under our care recently a rasping, systolic, aortic murmur could be clearly heard by a group of students standing at the foot of the bed."

Johnston (114) included a phonocardiographic tracing of an extracardiac sound heard at a distance, perhaps an example of pericardial knock.

Bellet and his associates have made a notable contribution to precordial noises in their articles on musical murmurs of aortic insufficiency (115, 116). They emphasized the part played by eversion of an aortic cusp diseased by syphilis. In their first paper only one of the 11 patients had murmurs heard at a distance. "The patient's bedfellow was considerably annoyed by the unusual and constant noise." In this and three other instances the murmurs were audible to the patient, presumably in the same distressing manner as Stokes' poor gentleman who likened himself to a humming top. This distinction between murmurs heard by a patient but not by others suggests that vibrations may be transmitted directly to the cochlear apparatus through the bone, blood vessels or other tissues of the body, rather than by air transmission. In their second group of patients four of 18 patients complained of hearing noises which interfered with sleep and made them nervous. One man produced such a noise that it disturbed his wife at a distance of seven feet.

Nichols (117) in his review of syphilis of the aorta has the following observations which suggest possible rupture of an aortic valve scarred by the lesion of syphilis.

"In five cases a peculiar musical diastolic murmur was heard, which seems worthy of comment. Musical murmurs are usually heard over the base of the heart in the aortic area. They are most often diastolic, rarely systolic, in time The usual explanation given for such musical diastolic murmurs is a rupture of a valve segment, perforation of a valve segment, or a cordlike strand of tissue across the valve orifice These five cases showed all the signs of aortic incompetence, in addition to this unusually loud high-pitched musical diastolic murmur Its very loudness suggested something moving freely back and forth in the blood stream The murmur was variable in intensity and exaggerated by exercise It was loudest over the aorta, but widely transmitted over the chest and, in three cases, was heard distinctly in the back It was distinctly decrescendo in type and usually filled the entire diastole In each case the patient was conscious of this 'whizzing' or 'singing' noise in his chest, and in three of the cases it was sufficiently loud to be audible to the unaided ear of the examiner at a distance of from one to three feet from the chest wall It was in all cases accompanied by a thrill, diastolic in time, most marked over the upper sternum and transmitted widely to the whole chest wall, but not to the soft structures of the neck That this murmur developed its intensity rather suddenly is attested by the fact that in three cases it occurred after a hard day's work and, in at least two and probably three cases, marked the beginning of myocardial failure"

Scott (118) commented on several instances of loud musical aortic diastolic murmurs heard at a distance in eversion of an aortic cusp as a sequel of syphilis.

Baer, Taussig and Oppenheimer (119) recorded a loud systolic murmur heard two inches from the chest in a patient with arachnodactyly who had an aneurysmal dilatation of the ascending aorta

Scherf and Boyd (120) discuss the condition as follows:

"Traumatic aortic insufficiency is also rare It occurs after direct and indirect cardiac trauma, irrespective of whether the valves are normal or abnormal. Sudden

at some distance from the chest wall by the unaided ear. The murmur is usually accompanied by a thrill Immediately after the provocative exertion, severe pain is felt over the heart and midsternal region, as the result of acute cardiac dilatation and stretching of the pericardium This form of aortic regurgitation, like the other types, is more common in males It developed in one of our patients during a brawl, and in another while playing soccer Both patients had syphilitic aortitis."

Kissane, Koons and Clark (121) reported two patients with traumatic rupture of the aortic valve. One, a 58-year-old carpenter, tripped and fell on his chest. "He immediately arose and walked to his automobile where he felt and heard a thrill or purring in his upper chest and noticed soreness in the sternal region. As he drove home he continued to hear this peculiar sound and developed some palpitation, tachycardia and slight dyspnea. That evening his wife was alarmed when she heard this purring-like noise at a distance of three feet from the patient " Aortic regurgitation was found, and after several bouts of congestive failure he died 27 months after injury. At autopsy "the aortic valve presented an unusual picture in that there was a splitting and separation of the commissures between the right and left posterior cusps, resulting in a sagging, free flap-like part of these cusps which moved with equal ease either upward into the aorta or downward into the left ventricle and produced a definite, permanent opening along the line of the separated commissures "

Levine and Harvey (122) have commented on murmurs of aortic stenosis which may be so loud as to be heard by the unaided ear at a distance from the chest. This super murmur was graded VI, the top of the scale.

While this section was being written I saw a patient with rupture of the aortic valve on the Medical Service of the University Hospitals at Iowa City. He was a cement worker, 41 years old. He and several members of

exercise and some other spells of palpitation with tachycardia they had not interfered with work. There was no story of rheumatic fever. When he

was examined for the army in 1941 nothing was found wrong with the heart but he was disqualified because of his eyes. On 19 September, eight weeks before hospitalization, he experienced the sudden start of dyspnea while he was putting a cement floor in a hog house; but there was no direct connection with any act of lifting or straining even though he joked about handling 100 pound objects as trivial Dyspnea even at rest has characterized his state ever since. Orthopnea and edema increased in severity till he was admitted to the hospital.

Examination disclosed the usual features of congestive failure and the peripheral and cardiac signs of free aortic regurgitation. After his condition improved with a regimen of rest, digitalis, diuretics and restriction of salt he was much improved. The heart evidenced great overactivity chiefly of the left ventricle. A diastolic thrill was very intense in the second right interspace and the third and fourth left interspaces. Pulsation of the subclavian and carotid arteries bespoke the wide pulse pressure with the reading 150/0 mm. Hg. There were pistol shots, Duroziez's murmurs, Corrigan pulse and capillary pulsations At the base there was a loud diastolic murmur with a rough grating quality but not at all musical. This spread all over the precordium, and over the mitral area I heard the presystolic rumble of an Austin Flint murmur, which had been mistaken for that of mitral stenosis on admission. A loud systolic murmur occurred at the base. There was no aortic valve closure sound but P_2 was loud. The diastolic murmur was so loud that it was clearly heard by the unaided ear an inch from the chest on the ward and six inches away in a quiet room It was not audible to the patient X-ray revealed 4+ left ventricular enlargement and no left auricular enlargement. Electrocardiograms exhibited the pattern of left ventricular hypertrophy After further convalescence he was discharged; and though his exercise tolerance was poor he was not in congestive failure He died rather suddenly four weeks after he went home, symptoms of failure having returned and increased in severity

The latest reference to such noises is that of Schiødt (123) who heard a systolic murmur 15 centimeters from the chest in a 58-year-old farmer with asymptomatic aortic stenosis The examining physician first heard the noise while examining the clothed patient with the ophthalmoscope!

BIZARRE AND MISCELLANEOUS SOUNDS

Schiødt has called attention to a strange case recorded by Bartholin in 1654 of "a young lady of Copenhagen sometimes vexed by a continuous headache. At these times she felt the pulse in the carotid arteries in her head so violently that a sound might be heard from a long distance, like a clock." It is not certain that this came from the precordium but is recorded as a curiosity. Another strange case is the one in a footnote in Laennec's

book on ausculation (5), a case recorded by Andral in the following terms: "I lately saw a woman who complained of palpitations of the heart. Each stroke of this organ was accompanied by a peculiar gurgling sound, which evidently came from the precordial region, and was heard only when the heart struck the ribs; it was perceptible at a distance."

Perhaps some of Laennec's should be included here and some of the Muhlengerausch of German authors late in the last century.

Frost and Bing (34) whose admirable but brief review of the subject of loud precordial noises appeared in 1940, described a healthy 22-year-old woman who was seized suddenly by a "crick in the back", a sensation that something had come loose in her chest and then pain in the chest and left shoulder. A blowing systolic murmur and some vague scratching sounds were heard along the left sternal border. During hospitalization she had a sense of precordial fullness, "like a ball being squeezed out between two fingers" and sounds came from her precordium so loud that they were heard 3 meters away. The sounds were synchronous with the heart and loudest when she was lying on her left side, disappearing when she turned to the right. Phonocardiograms showed the sounds to be systolic in time but spaced at variable intervals from beat to beat. Intensive x-ray and other studies failed to reveal any lesion of lung, pleura, pericardium or heart The likeliest guess is that pulmonary interstitial emphysema was responsible but the issue is in doubt.

Warburg, who scarcely mentioned such phenomena in his monograph on cardiac trauma (15), reported elsewhere (124) a man with mitral stenosis and auricular fibrillation observed many times in congestive heart failure. With an obscure infection he had a short fever and "during the last 24 hours he had heard a sound from his chest which he described as though something were dripping; he said, that he thought his heart had burst. His wife was able to hear the sound when she was lying in the bed beside him. I was able to verify his statements At every heart beat a clicking or slightly sonorous sound was audible in the room. . A phonocardiogram showed that besides the sounds usually found in this patient there was a new murmur, partly during the systole, partly immediately after the systole It was quite clear that the new sound included higher frequencies than the other sounds. The phonocardiogram and the electrocardiogram taken are seen as the lower two curves of the figure " No cause was suggested.

Levine and Harvey (122) recorded the case of a 45-year-old woman with well compensated mitral stenosis and auricular fibrillation. After exercise a weird-sounding, rough musical murmur "was actually heard with the naked ear a foot away from the chest For several years she had been aware of a peculiar noise in her chest at times We have heard it at irregular intervals, sometimes only for a few seconds, at other times constantly and

then it might be absent for days. It is not related to position of the body or to breathing.... There is no x-ray evidence of diaphragmatic hernia or any other abnormality that might throw light on its causation."

Beaumont (125) mentions parenthetically the case of a "Mrs. W. complaining of noises over the heart which kept her awake. I found no evidence of pericarditis but loud rhonchi were audible over the lungs." Its nature remains obscure.

To wind up this amorphous group I append two casual notes. Dr. Lewis January (126) has called to my attention a rare instance of a healthy young man whose heart sounds were clearly audible several inches from his mouth when it was held open. He had no heart disease. X-ray study of the chest, esophagus and upper G.I. tract was normal. Perhaps some anatomical quirk permitted his esophagus to be patulous when his mouth was open and to act as a megaphone. Though I have looked for such a sign many times I have not yet come across it.

I conclude with the following autobiographical note: Occasionally when my stomach contains just the proper quantity of fluid and air, a happy postprandial mixture, the exact amount of each I have never been able to discover, and when semi-recumbent in an easy chair a systolic tinkle or splash is clearly audible to me and very diverting. It has been heard by others a foot away. The cardiac impulse on the diaphragm and stomach obviously produces the sound. It generally vanishes after a few minutes or can be eliminated by belching; but swallowing air or air and water has not brought it back at will.

DISCUSSION

Beyond serving as a repository for esoterica, is there usefulness in collecting together such diverse disorders with the common denominator of noise heard at a distance from the chest? Certain gleanings may reward closer study. While the figures defy statistical analysis, they suggest that loud heart murmurs are the commonest cause of the phenomenon, followed by interstitial emphysema, pneumothorax and pneumopericardium (table 1). Records of the rest of the conditions are less numerous. From the topographical and mechanical view air loose in the chest from the interstitial emphysema or pneumothorax, spontaneous or accidental, heads the list Cardiac murmurs come next and then pneumopericardium Since the sources of this review have been casual and the survey sporadic, the numbers may not be representative In some specialties particular problems may be missing or too heavily represented A surgeon's casual reading and experience would no doubt assemble a different array of noise-producing conditions. The cases included in this review are summarized in table 1.

The general problem of such noises may be considered by using interstitial

emphysema of the lung as an example. In such a disturbance the noises heard at a distance are an exaggerated sign which much more often is confined to the chest and can be heard only by the customary method with

TABLE 1

Conditions Causing Precordial Noises

	NUMBER	PER CENT	TOTAL PER CENT
Condition			
Heart murmurs	57	35	35
Spontaneous interstitial emphysema	28	17 }	21
Traumatic interstitial emphysema	6	4 }	
Spontaneous pneumothorax	15	9 }	16
Traumatic pneumothorax	12	7 }	
Pneumopericardium	25	15	15
Alimentary canal	8	5	5
Miscellaneous	7	4	4
Air embolism	6	4	4
Location			
Lung and chest	61		37
Heart valve	57		35
Pericardium	25		15
Alimentary canal	8		5
Miscellaneous	7		4
Air embolism	6		4
Organ			
Heart			
Valve	57 }		
Pericardium	25 }		55
Air embolism	6 }		
Chest and lung	61		36
Other	15		9
Mechanism			
Air	100		61
Heart valve	57		35
Other	7		4

irregular timing of such noises during systole, and perhaps quiet the suspicion of skeptics that such noises are illusions What constitutes the urge to publish case reports has never been known, but one has only to hear

these eerie sounds to appreciate the high clinical drama. Published cases reflect an unknown fraction of those met clinically. Certainly luminous papers and talks, such as the late Louis Hamman produced, were followed by a wave of case reports, and a useful diagnostic sign caught popular fancy and became a medical fashion.

Clinical features of spontaneous interstitial emphysema of the lung may suggest acute myocardial infarction, but usually the complaint of pain in the chest is more insistent than the meager signs of difficulty. The patient is in pain but looks well, usually breathes easily and is not in shock. Generally he is young, vigorous and active, without sign or story of hypertension, angina or vascular disease. Later reactions such as fever, leukocytosis, fast blood sedimentation do not follow. If the patient has a loud noise it may be produced only in certain positions so that change of position or movement can enhance or quiet the ticking chest Table 2 contains some of the data relating to the noises which have been reported as audible at a distance, a feature of about 10 per cent of the recorded cases. The loudest noise, measured in distance heard, was perceived 20 feet from the patient. Duration varied from 2 hours to 2 weeks. There was much variation in the intensity; sometimes everything would be quiet and then the sounds returned. As a general rule they would come and go. In all except one, the sounds were heard best with the patient lying on his or her left side, and many were able to demonstrate the sound at will by assuming the proper position.

I have seen one young man (the notes of his case have vanished) whose main distress was not the pain in the chest, but that the noise coming from his chest was so loud his wife made him sleep in the next room. He could not turn and eliminate it but finally it went away and he gained his reprieve Several other reports suggest that such noises are a rare cause of temporary domestic infelicity; and they are alarming to patient and family. Indeed fear and curiosity occasionally have brought the patient to the doctor, since there may be very little pain.

Subcutaneous emphysema has been recorded in only a few cases (table 2) but since it may be confined to a small area and be ephemeral it is easily overlooked

In practically all cases of interstitial emphysema of the lung, if complicating pneumothorax occurred it was left-sided. Often it has been so small that only careful search with proper alignment has produced diagnostic x-ray shadows. Right-sided pneumothorax does not appear to be rarer than the left-sided kind, but only uncommonly has been found in association with the crunching, bubbling, paper-rustling sound (Hamman's sign) so characteristic of mediastinal emphysema. I found no record of right pneumothorax in conjunction with the distantly heard noise.

The most comprehensive discussion of such sounds in interstitial emphy-

sema was given by Greene (43) who differentiated between two classes of sounds. "The bubbling, crunching, clicking and some of the tapping sounds are due to the heart rubbing against emphysematous blebs in interstitial

TABLE 2

Interstitial Emphysema of the Lung

AUTHOR	DISTANCE HEARD	DURATION	CONSTANCY	POSITION CAUSING LOUDEST NOISE	PNEUMOTHORAX	SUBCUTANEOUS EMPHYSEMA
Hamman	Several feet	Few days	—	On left side	0	0
	One foot	8 days	0	On left side and leaning forward	0	0
	Several feet	2 days	Recurrent	Flat on back	Small left	0
Morey & Sosman	Several feet	2 weeks	Recurrent	Left side	Small left	0
McGuire & Bean	Several feet	4 days	Intermittent	Left side	Small left	+
Wolff	One foot	7 days	Intermittent	Left side	Small left	0
Frost & Bing	10 feet	—	Intermittent	Left side	0	0
Styron	Several feet	Few hours	Intermittent	Left side	0	0
Pinckney	20 feet	Few days	3 times mos apart	Left side	Left	0
	Several feet	24 hours	—	—	—	—
Caldwell	Many feet	7 days	Intermittent	Left side	0	0
Meek	Several feet	Several weeks	—	Left side	—	—
Griffin	Several feet	Several days	Intermittent	Semirecumbent	Left	0
	Heard by patient	5 days	—	—	Left	—
Lintz	Several feet	2 hours	—	—	Small left	+
Greene	Several feet	10 days	—	Left side	Air in mediastinum	0
McCabe, 4 cases	—	2–5 days	—	—	Air in mediastinum & left pneumothorax	0
Dickie	Heard by patient	Several days	—	—	Left	0
	Several feet	—	—	—	Air in mediastinum	0
	Heard by patient	Few hours	—	Left	Small left	0
	—	—	—	—	Small left	0
Aisner & Franco	Heard by patient	—	Intermittent	—	0	0

emphysema of the lung and mediastinum The knocking and tapping

human experiments is the best we are likely to get from observing the effects of accidents and disease.

While the absence of distantly heard noises can only be inferred, the consistency of the record suggests that there is clinical significance in its omission from certain reported cases. For instance in the large literature of pneumoperitoneum such noises are not listed. I have not encountered the phenomenon in reading about or seeing the abdominal catastrophes in which air leaks from the ruptured gut. It is not recorded in reports of interstitial emphysema and left-sided pneumothorax in infants and young children although Hamman's sign occurs. The assumption is that the noises require the larger heart and fuller sounding board of the adult thorax for their distant propagation

Diagnosis of such rare conditions is important because the cause may be a mechanical crisis which can be corrected or eased Delay or confusion may be fatal On the other hand, if the causal condition is innocuous, it is well to avoid mistaking it for its clinical counterfeits of ominous import. In order of urgency the Mühlengeraüsch or mill wheel sound of air embolism stands first. Its clinical debut, always unexpected, is associated with an opening by wound, scalpel or needle by which air gets into veins. Immediate rotation to the left lateral position, or the Trendelenburg position, trapping air bubbles above the blood in the right ventricle where they may be aspirated with a needle and syringe may be life saving (95) Inhalation of 100 per cent oxygen may help.

Rupture of the esophagus, which I have seen produce Hamman's sign, probably can cause loud noises, even without pneumopericardium. Since surgical cure in such a calamity may now be anticipated, the diagnosis of a ruptured gullet should be considered especially where a story of extreme vomiting or instrumentation makes the condition probable

Traumatic pneumothorax, pneumopericardium, and pneumomediastinum have obvious urgency with the connotation of crushed ribs, or penetration with missile or foreign body The attending noise may indicate the extent of damage which otherwise is not apparent. Treatment is that of the underlying condition

Spontaneous interstitial emphysema of the lung has the implication of pneumothorax and at times the hazard of tuberculosis The noise heard at a distance has not been nearly so ominous as it has seemed to the victim, always amazed and often terrified by such uncanny behavior Thus diagnosis usually permits reassurance, since all recorded cases have survived A note of warning should remain us of tension pneumothorax which may lurk under this clinical camouflage. Also "air lock", the dissection of air through the lung or hilum under enough pressure to impede blood flow, may require more heroic measures, 100 per cent oxygen, or perhaps operation to stop

the leak if it can be found. In general the great role of the physician here is to calm terror by bringing assurance.

Noise from the esophagus may rarely lead to the discovery of hiatus hernia with an errant stomach or colon.

Loud murmurs, when they have arisen out of the innocuous quiet of the past, especially under some stress or accident, may call to mind the likelihood of ruptured aortic valve, or the turning inside-out of a syphilitic valve cusp Giving a better view of prognosis, such diagnostic *tours de force* escape the odium of academic banality.

Pneumopericardium, apparently of great rarity now, calls for a review of the eroding and destroying processes which permit air to escape into the pericardium from adjoining air-containing structures. Therapeutic pneumopericardium has its own obvious cause; and its insubstantial vogue is past.

SUMMARY

1 A hodge-podge of clinical conditions which may be associated with precordial sounds heard by the unaided ear at a distance from the chest has been assembled, compared and assessed

2. The commonest cause of such sounds was cardiac murmurs produced by valve rupture or other lesion, often abrupt in onset, and a consequence of stress or strain Next in order of frequency came interstitial mediastinal and pulmonary emphysema, both the spontaneous and traumatic This was followed by spontaneous and traumatic pneumothorax, pneumopericardium, noises the heart produced by striking air-containing gut, air embolus, and a small mysterious miscellany of unexplained sounds.

3 Since the noises here considered have many diverse sources there is no patho-physiologic common denominator to compare with the clinical fact of abnormally great volume of sound With such difference in cause and like difference in necessary treatment and in prognosis attention should be given to the cause, which usually comes to light upon clear clinical scrutiny.

REFERENCES

Introduction

1 McGuire, J and Bean, W B Spontaneous interstitial emphysema of the lungs Am J Med Sc, 197: 502, 1939

[illegible] al

[illegible] n,

1: [illegible], 1940

4. Paulley, J. W, Lees, D H and Pearson, A C Cough fracture of late pregnancy Brit Med J, 1: 135, 1949

5 Laennec, R. T. H. De l'auscultation médiate, Brosson et Chaudé, Paris 1819, Forbes Translation of Ed 3 Samuel and William Wood, N Y, 1838

Traumatic Pneumothorax

6 BRICHETEAU Observation d'hydro pneumo-péricarde, accompagnée d'un bruit de fluctuation perceptible à l'oreille Arch. Gén Méd., 4th series, 4: p 331, 1844

7. MOREL-LAVALLÉE: Rupture de péricarde bruit de roue hydraulique: bruit de moulin. Gaz. Med Paris, 19: 695, 729 & 803, 1864 [Acute early observations and much speculation.]

8 REYNIER: Recherches cliniques et expérimentales sur le bruit de moulin, symptome d'épanchement intra et extra péricardique dans les traumatismes de la poitrine. Arch. Gen de Med, 145: 441-582, 1880

9 MORRIS, H. C, L A remarkable case of pneumothorax Lancet, 2: 1121, 1902

10 REES, W. A AND HUGHES, G S: Wounds of the chest Lancet, 1: 58, 1918

11 SMITH, S M.: Pericardial knock Brit Med J., 1: 78, 1918

12 MUNDEN, W P. H., Memoranda Brit. Med J, 1: 174, 1918.

13 BLANKENHORN, M. A. From discussions and class notes

14 SPECHT, O.. Ein Fall von Mühlengeräusch nach Brustquetschung München med Wchschr, 67: 1118, 1920

15 SCHILLING, quoted by Thiem, in Warburg, E Subacute and chronic pericardial and myocardial lesions due to non-penetrating traumatic injuries, p 36 Levin & Munksgaard, Copenhagen, 1938

16 FUNK, D C: Personal report

Spontaneous and Induced Pneumothorax

17. CORNILS Ein Fall von Pneumothorax mit par distance Hörbaren Herztöner Deut Med Woch, 11: 1885.

18 ALBERT, A. Über das Auftreten des Mühlengeräusches des Herzens, als Zeichen einer Luftembolie ins rechte Herz bei Pneumothorax-behandlung Beitr z Klin d Tuberk, 52: 284, 1922

19 BOYÉ Beitr z Klin d. Tuberk, 52: 296, 1922 (discussion of Albert)

20 JUNKER, F Mühlengeräusch als komplikation beim kunstlichen Pneumothorax Beitr z Klin d Tuberk, 63: 497, 1926

21. LISTER, W A A case of pericardial knock associated with spontaneous pneumothorax Lancet, 1: 1226, 1928

22 WOLFERTH, C C AND WOOD, F C Angina pectoris Med Clin North Am, 13: 947, 1930

23 SCADDING, J C AND WOOD, P Systolic clicks due to left-sided pneumothorax Lancet, 2: 1208, 1939

24 SHARPEY-SCHAFER, E P Letter Lancet, 1: 237 (1388) 1940

25 Discussion Quart J Med, 8: 381, 1939

26 EDWARDS, P W AND SIMPSON, T Observations on "pericardial knock" Tubercle, 20: 426, 1939

Interstitial Emphysema of Lung and Mediastinum

27 PETERSEN, G: Ein Fall von extrapericardialem emphysem Berlin, klin Wehnschr, p 699, 1884

28 EDELFSEN: Quoted by Petersen

29 HAMMAN, L Remarks on the diagnosis of coronary occlusion Ann Int Med, 8: 417, 1934

30. HAMMAN, L Spontaneous interstitial emphysema Trans Assoc Am Physicians, 52: 311, 1937, [The classic paper]

31. Hamman, L.: Spontaneous mediastinal emphysema. Bull. Johns Hopkins Hosp., 66: 1, 1939
32. Morey, J B and Sosman, M. C.: Spontaneous mediastinal emphysema Radiology, 32: 19, 1939.
33. Wolff, B P.: Spontaneous interstitial emphysema of the lungs; report of an additional case Ann Int. Med, 13: 1250, 1940
34 Frost, J. and Bing, J. Some cases of precordial sounds audible at a distance Acta Med. Scand, 105: 411, 1940
35 Styron, C. W.: Spontaneous mediastinal emphysema. New Eng J. Med, 225: 908, 1941.
36. Pinckney, M M Mediastinal emphysema and idiopathic spontaneous pneumothorax Va. Med Mo, 68: 315, 1941.
37. Caldwell, H W: Spontaneous mediastinal emphysema J A M. A, 116: 301, 1941.
38 Miller, I Spontaneous interstitial emphysema of the lungs: a report of two cases and summary of the literature Ohio State Med. J, 37: 1056, 1941.
39. Meek, E M: Spontaneous mediastinal emphysema South Med J., 35: 990, 1942.
40 Griffin, R. J.. Spontaneous pneumothroax. Ky. Med J, 39: 284, 1941
41. Griffin, R J: A diagnostic sign of spontaneous intersitial emphysema of the mediastinum, case reports Ann. Int. Med., 17: 295, 1942
42 Lintz, R M Spontaneous mediastinal emphysema Arch Int Med, 71: 256, 1943
43 Greene, J A: Unusual sounds emanating from the chest Arch Int Med, 71: 410, 1943
44 Miller, H Spontaneous mediastinal emphysema Ann Int Med, 21: 998, 1944.
45 McCabe, E. S: Spontaneous interstitial emphysema of the lung simulating organic heart disease Am Heart J, 34: 729, 1947.
46 Dickie, H A: Spontaneous mediastinal emphysema and spontaneous pneumothorax—a report of 20 cases Ann Int Med, 28: 618, 1948

in the light of laboratory experiment Medicine, 23: 281, 1944.
48 Aisner, M and Franco, J E Mediastinal emphysema New Eng J Med, 241: 818, 1949
49 Torrey, R G and Grosk, L C Acute pulmonary emphysema observed during the epidemic of influenzal pneumonia at Camp Hancock, Georgia Am J Med Sc, 157: 170, 1919
50. Clark, E and Synnott, M J Influenza pneumonia cases showing gas in *fascial tissues* Am J Med Sc, 157: 219, 1919
51 Berkely, H K and Coffen, T H Generalized interstitial emphysema and spontaneous pneumothorax as complications of bronchopneumonia J A M A, 72: 535, 1919
52 Bullowa, J G M Tissue emphysema in influenza Med Rec, 95: 346, 1919
53 Gordon, C. A.: Respiratory emphysema in labor Am J Ob & Gyn, 14: 633 1927
54. MacRae, D J.. Spontaneous pneumomediastinum in pregnancy Lancet, 1: 902, 1949.

Traumatic Mediastinal Emphysema

55 STEIGER, Quoted by James (81): Würtumberg med. Zeit., 1864.
56 LEONSPACHER, Quoted by James (81): Aerzt. intell Blatt., 1875
57. WEIL, S: Mediastinalemphysem mit Muhlengeraüsch nach Plexus anästhesie Zentralblatt für Chir., 46: 890, 1919.
58 JEHN, W.: Das Mediastinalemphysem. Zentr. f. Chir., 482: 1619, 1921.
59 HÖRNICKE, C. B., Ueber das sogenannte Mühlengerausch Munchen med Wchnschr., 69: 819, 1922.
60 BIGGER, I. A.: Wounds of the heart and pericardium South. Med J., 25: 785, 1932

Alimentary Canal

61 BOERHAAVE, H.: Commentaries upon the aphorisms of Dr. Herman Boerhaave by Van Sevieten, G. Horsfield and Longman, London, 2: 112, 1744
62 BEGBIE, J W.: Half Yearly Abstract of Medical Sciences, 1: 117, 1863
63 ALLEN, F. N. Heart noises. New Eng Med. J., 218: 655, 1938
64 RYLE, J. A. The Natural History of Disease. Ed 2, Oxford University Press, 1948
65 ROBERTS, J T · Dynamics and circulation of heart muscle, cardiac reserve, and the cardiac cycle In Sodeman, W A, Pathologic Physiology, p 48 W B Saunders Co, Phila, 1950

Heart

66 MORGAGNI, J. B The seats and causes of diseases Translated by Alexander, B, London, Miller & Cadell, 1769
67. PORTAL, Quoted by Brichteau (68)

Pneumopericardium

68 BRICHTEAU Observation d'hydro-pneumo-péricarde, accompagnée d'un bruit de fluctuation perceptible à l'oreille Arch Gén Méd 4th series, 4: 334, 1844
69 STOKES, W Diseases of the Heart and the Aorta Lindsay & Blakiston, Philadephia, p 37, 1855
70 ARAN, Quoted by Gibson, G A Diseases of the Heart and Aorta, p 376, Pentland, Edinburgh, 1898
71 NIEMEYER Practice of Medicine, 1: 393 Translated by Humphrey & Hackley, New York, 1862 [See also Deut klin, 1860]
72 EISENLOHR, C Ein Fall von Pyopneumopericardie Berliner klin Woch., p 473 1873
73 FRIEDREICH, Quoted by James
74 FETZER, Quoted by James Wurten Aerzte ver, 1874
75 MEIGS, J F Case of pneumo-hydropericarditis, with remarks. Am J Med Sc, 69: 81, 1875
76 GUTTMANN, P Pneumopericardium, entstanden durch Perforation eines runden Magengeschwurs in den Herzbeutel Berlin klin Woch, 221, 1880
77 LOVE, J K Case of hydro-pneumo-pericarditis Lancet, 1: 319, 1888
78 NICHOLLS, A G Notes on some cases of infection by the bacillus aerogenes capsulatus Brit Med J, 2: 1844, 1879
79 LAUB, M Ein Fall von pneumopericardium Wein klin Woch, 156 (No 7), 1899

80 Saexinger, Quoted by James· Prager Med. Woch , 13, 1901
81 James, W. B.: Pneumopericardium. Trans. Assoc Am Physicians, 19: 351, 1904.
82 Meyer, A.: A case of spontaneous pyopneumothorax complicated by hydro or pyopneumocardium. Med Record, 88: 991, 1915.
83 Yates, W. N.· Injury with extravasation of blood in the pericardium. J. Missouri St. Med. Assoc , 13: 29, 1916.
84. Heise, F. H. and Brown, L : A case of hydropneumopericardium in a tuberculous individual during an attack of typhoid fever. Am Rev Tuberc , 8: 284, 1923
85 Stahl, R. and Entzian, W. Klinisches und Experimentelles uber das intra- und extrakardiale muhlengrausch Zeit fur klin. Med , 100: 232, 1924.
86 Shackleford, R T : Hydropneumopericardium J A M A , 96: 187, 1931.
87. Gilbert, A. A Spontaneous pneumopericardium· with report of a case J Arkansas Med Soc., 35: 53, 1938
88 Trimble, H G , Eaton, J L and Thompson, K : Spontaneous pneumopericardium in a case of apparently arrested tuberculosis Am. Rev Tuberc , 45: 100, 1942.
89 Price, F W A Textbook of the Practice of Medicine, 8th Ed. Oxford Med Publications, 1950.

Air Embolism

90 Wagner, J · Ueber das Mühlengeraüsch des Herzens München med Wochnschr, 692: 1543, 1922
91 Hirsch, E and Sauser, G Muhlengeraüsch als Symptom einer Luftembolie des rechten Herzens beim kunstlichen Pneumothorax. Wien klin. Wchnschr. 48: 232, 1935
92 Durant, T M , Long, J and Oppenheimer, M. J. Pulmonary (venous) air embolism Am Heart J , 33: 269, 1947
93 Crile, G , Jr Practical Aspects of Thyroid Disease, p 170 W B Saunders, Philadelphia and London, 1949
94. DeGowin, E L , Hardin, R C and Alsever, J B Blood Transfusion, p. 291 W. B Saunders, Philadelphia, 1949
95 Stallworth, J M, Martin, J B and Postlethwait, R W Aspiration of the heart in air embolism J A M A , 143: 1250, 1950

Heart Murmurs

96 Burns, A . Observations on some of the most frequent and important diseases of the heart, pp 187–189 Thomas Bryce, Edinburgh, 1809
97 Corvisart, J N Essai sur les maladies et les lésions organiques du coeur, Ed 3. Mequignon-Marvis, Paris, 1818
98. Graves, R L Clinical lectures London Med & Surg J , 7: 516, 1835
99 Latham, P M Diseases of the Heart, p 37 Barrington & Haswell, Philadelphia, 1847.
[illegible]

subsequent reviews.
101. Peacock· Case of diseased heart in which a musical murmur was heard Trans Path Soc. London, 6: 55, 1854
102. Simpson, H.: A case of aortic regurgitation from injury to the valves during muscular effort Brit Med. J., 2: 167, 1868.

103. Yeo, J. B.: Rupture of the aortic valves. Lancet, 2: 792, 1874.
104. Orton: Case of rupture of the aortic valve. Lancet, 1: 10, 1877.
105. Frew and Finlayson: Rupture of aortic valve typical history. Brit. Med. J., 1: 936, 1879.
106. Barie, E.: *Recherches cliniques et expérimentales sur les ruptures valvulaire du coeur.* Rev. de méd., 1: 132, 1881.
107. Tretzel, L.: Ruptur einer Aortenklappe in Folge körperlicher Anstrengung. Berlin klin. Woch., 28: 1073, 1891.
108. Langwill, H. G.: An unusual cardiac case. Scottish Med. Surg. J., 1: 723, 1897.
109. Marshall, A. L.: Case report. Lancet, 1: 1079, 1908.
110. Allbutt, T. C.: In System of Medicine, Ed. 2, Vol. 6, p. 447. Allbutt, T. C. & Rolliston H. D. Macmillan & Co., London, 1909.
111. Hoffmann, A.: *Herz und Gefäßkrankheiten und Unfall.* Med. klin., 7: 1569, 1912.
112. Wilson, F. N. and Jamieson, R. A.: Musical diastolic murmurs in aortic insufficiency. Heart, 7: 71, 1919.
113. Major, R. H.: Physical Diagnosis, Ed. 1, p. 232. W. B. Saunders, Philadelphia, 1937.
114. Johnston, F. D.: *Extra sounds occurring in cardiac systole.* Am. Heart J., 15: 221, 1938.
115. Bellet, S., Gouley, B., Nichols, C. F. and McMillan, T. M.: Loud musical diastolic murmurs of aortic insufficiency. Am. Heart J., 18: 483, 1939.
116. Gelfand, D. and Bellet, S.: The musical murmur of aortic insufficiency. clinical manifestations based on a study of 18 cases. Am. J. Med. Sc., 221: 644, 1951.
117. Nichols, C. F.: A study of syphilis of the aorta and aortic valve area. Ann. Int. Med., 14: 960, 1940.
118. Scott, R. W.: Cardiovascular syphilis. Oxford Med., 2 (Part 2): 508, 1942.
119. Baer, R. W., Taussig, H. B. and Oppenheimer, E. H.: Congenital aneurysmal dilatation of the aorta associated with arachnodactyly. Bull. Johns Hopkins Hosp., 72: 309, 1943.
120. Scherf, D. and Boyd, L. J.: Cardiovascular Diseases. J. B. Lippincott, Philadelphia, 1947.
121. Kissane, R. W., Koons, R. A. and Clark, T. E.: Traumatic rupture of the aortic valve. Am. J. of Med., 4: 606, 1948.
122. Levine, S. A. and Harvey, W. P.: Clinical Auscultation of the Heart. W. B. Saunders Co., Philadelphia & London, 1949.
123. Schødt, E.: Heart sounds audible at a distance, a case described in 1654 and one in 1951. Acta Med. Scan. 266 (Sup.) 887, 1952.

Miscellaneous

124. Warburg, E.: Cardiac murmurs audible at-a-distance—mitral stenosis—thrombosis of the right ventricle—pulmonary infarction. Acta Med. Scand., 107: 503, 1941.
125. Beaumont, G. E.: Applied Medicine, p. 158. Blakiston Co., Philadelphia, 1950.
126. January, L. E.: Unpublished observations.

The Physiology of the Body Fluids

WILLIAM M. WALLACE, M.D.

The cellular metabolic operations essential to life are carried out in dilute aqueous solutions of inorganic salts. The concentrations of the various salts and the volume of the body fluids are highly critical factors for proper cellular function. A great part of the functional energy of the heart, kidneys and endocrine system is devoted to the task of maintaining the constancy of composition and volume of the body fluids in the face of the constant hazard of a dry and salt-free external environment. The description of the mechanisms by which this constancy is achieved constitutes the subject matter of body fluid physiology.

Progress in the use of antibiotic substances and in surgical technique has greatly extended the critical periods during which the clinician must support the volume and composition of the body fluids In addition, the extensive use of salt-affecting hormones and of low-salt diets has increased the complexity of the problems faced by the clinician. A thorough knowledge of normal and abnormal physiology of the body fluids has become essential to adequate management of many patients. The parenteral administration of fluids is the most commonly used life-saving procedure in medical practice.

The present discussion is intended to furnish a resume of the clinical physiology and pathology of the body fluids. More rigorous and detailed discussion are to be found in the monographs of Gamble (1), Peters and Van Slyke (2), Peters (3), and Smith (4) Shorter reviews have been presented recently by Butler and Talbot (5), Darrow and Pratt (6), Elkinton (7) and by Gamble (8). Excellent clinical discussions are those by Marriot (9) and Newburgh (10).

DEFINITION OF TERMS

Progress in the understanding of body fluid physiology in the past 25 years, in large part, has grown out of the successful application of the electrolytic dissociation theory to the subject Familiarity with this concept and its terminology is essential for an understanding of the field and for its clinical application. This concept of the behavior of dissolved substance in water contains the following approximate assumptions:

1. An electrolyte dissolved in water dissociates into two or more particles called ions which bear opposite electrical charges. The electropositive (basic) particle is called the *cation*, the electronegative particle (acidic) is called the *anion*. The sum of anions and cations must always be equal in a solution.

2. Ions in solution behave like molecules in determining the osmotic pressure and other physical properties of the solution. Thus, sodium chloride dissociates into two ions and produces a solution with twice the osmotic pressure of a solution containing an equal number of molecules like urea or glucose (non-electrolytes).

3. The dissociation of an electrolyte is not complete, but involves an equilibrium between ions and the undissociated molecules of the electrolyte. The fraction of dissociation varies, having a small value for electrolytes like carbonic and phosphoric acid and a large value for ones like sodium chloride or bicarbonate. The tendency for the weak acids in a solution of electrolytes to dissociate hydrogen ions is expressed as the pH, acidity or reaction of the solution. Buffering, the ability of the body fluids to resist pH change, is a function of the equilibrium between weakly dissociated acids of the body fluids and the strongly dissociated salts of the fluids *Acid-base equilibrium* refers to the relative amounts of anions and cations functioning in this equilibrium An excellent discussion of the electrolytic dissociation theory as applied in medical physiology is found in Clark (11).

Earlier workers have expressed concentrations in terms of weight per volume, usually milligrams per 100 cubic millimeters. Since chemical reactions are most correctly interpreted in terms of ions or molecules rather than in terms of the actual weights of the substances, terminology based on chemical equivalence is more informative than that based on weight. The use of such terminology greatly facilitates the comprehension of the pathologic physiology of the body fluids For example, blood plasma contains 322 mg per cent of sodium and 350 mg. per cent of chloride—blood plasma would appear to contain more chloride than sodium. Actually the reverse is true in terms of equivalence, 140 meq. of sodium and 100 meq. of chloride are present. Since the sum of the equivalents in a solution like serum must be equal, attention is immediately focussed upon the difference between the two numbers If the serum sodium concentration is 140 meq. and the chloride is 120 meq it is immediately evident that the acid-base balance is greatly disturbed

The units used are defined as follows.

mol. (M) = molecular weight in grams.

molar solution = 1 mol made up to 1 liter with water.

equivalent, or *eq.* = 1 mol ÷ the valence Phosphate radicles are either univalent, divalent or trivalent depending upon the pH of the solution in which they occur. Hence the equivalence varies with the pH.

milli, or *m*, used as a prefix to mol or equivalent, written mM and meq., denotes 0.001 of the amount, and as a prefix to molar etc., denotes 0.001 of the concentration.

osmol, or *osM.* The amount of electrolyte equivalent in osmotic pressure to that of a molar solution of non-ionized solute. A molar solution of

The Physiology of the Body Fluids

WILLIAM M. WALLACE, M.D.

The cellular metabolic operations essential to life are carried out in dilute aqueous solutions of inorganic salts. The concentrations of the various salts and the volume of the body fluids are highly critical factors for proper cellular function A great part of the functional energy of the heart, kidneys and endocrine system is devoted to the task of maintaining the constancy of composition and volume of the body fluids in the face of the constant hazard of a dry and salt-free external environment The description of the mechanisms by which this constancy is achieved constitutes the subject matter of body fluid physiology.

Progress in the use of antibiotic substances and in surgical technique has greatly extended the critical periods during which the clinician must support the volume and composition of the body fluids. In addition, the extensive use of salt-affecting hormones and of low-salt diets has increased the complexity of the problems faced by the clinician. A thorough knowledge of normal and abnormal physiology of the body fluids has become essential to adequate management of many patients. The parenteral administration of fluids is the most commonly used life-saving procedure in medical practice.

The present discussion is intended to furnish a resume of the clinical physiology and pathology of the body fluids. More rigorous and detailed discussion are to be found in the monographs of Gamble (1), Peters and Van Slyke (2), Peters (3), and Smith (4) Shorter reviews have been presented recently by Butler and Talbot (5), Darrow and Pratt (6), Elkinton (7) and by Gamble (8) Excellent clinical discussions are those by Marriot (9) and Newburgh (10)

DEFINITION OF TERMS

Progress in the understanding of body fluid physiology in the past 25 years, in large part, has grown out of the successful application of the electrolytic dissociation theory to the subject Familiarity with this concept and its terminology is essential for an understanding of the field and for its clinical application This concept of the behavior of dissolved substance in water contains the following approximate assumptions:

1. An electrolyte dissolved in water dissociates into two or more particles called ions which bear opposite electrical charges. The electropositive (basic) particle is called the *cation*, the electronegative particle (acidic) is called the *anion*. The sum of anions and cations must always be equal in a solution

tem, kidneys, lungs, skin and gastrointestinal tract. Water and its dissolved salts shift freely across the capillary bed into the interstitial fluid. The balance between inflow and outflow at this point is a function of the hydrostatic pressure tending to drive fluid out and the colloid osmotic pressure of the plasma protein tending to restrain the outward movement. In certain specialized areas, such as the liver, these relations may be reversed

TABLE 1

Electrolyte Concentration in Extracellular Fluid (Normal Serum Values in meq./l. of Serum) and Intracellular Fluid (Muscle, meq./l. of Muscle Water)

CATIONS	BLOOD SERUM	MUSCLE WATER
Na	140 (136–145)	10
K	4 (3.5–5.0)	160
Ca	5 (4.5–5.3)	?
Mg	2 (1–2)	35
Total	151 (145–157)	205*
ANIONS		
Cl′	100 (98–104)	0?
HCO_3'	27 (26–28)	10
HPO_4''	2 (2–3)	120
SO_4''	1 (1–1.5)	20
Organic acid	4 (1–6)	?
Protein	17†	55
Total	151	205‡

* The greater total cation concentration in muscle water arises from the multivalency of protein and phosphate anions and does not indicate lack of osmotic equality. See Gamble (8) for discussion.

† Calculated for a normal serum protein concentration and A/G ratio at a serum pH of 7.45 from the data of Van Slyke and Assoc. (J. Biol. Chem., 79: 769, 1928).

‡ These are tentative and approximate values calculated from the literature.

by virtue of the increased capillary permeability to protein. The lymphatic channels may be considered an accessory route for exchange of extracellular fluid with the organs concerned with intake and excretion. This system is particularly concerned with the removal of protein that is continually lost in low concentration from the capillary bed. Although rigid proof is lacking, the preponderance of evidence indicates that the extra- and intracellular phases of the body are in osmotic equilibrium. Changes in the osmotic pressure of the compartments are immediately equalized by transfers of water. Much evidence indicates that selective permeability

sodium chloride is spoken of as being 2 osmolar or containing 2 osmols (2000 millosmoles).

Colloid osmotic pressure. This is often termed "oncotic pressure" and represents the fraction of total osmotic pressure of a solution than can be accounted for by non-diffusible ions in the solution. In blood plasma it is accounted for by the plasma proteins and, in terms of osmols, is only minute, approximately 0.1 per cent of the total osmotic pressure. Despite its small value, however, it is of major importance in determining the distribution of body water.

Conversion of milligrams per cent to millequivalents per liter is made by the use of the following relation:

$$\text{mEq per liter} = \frac{\text{milligrams per cent} \times 10 \times \text{Valence of ion}}{\text{molecular weight of ion}}$$

$$CO_2\ (\text{mEq/l.}) = \frac{\text{Vol } \%\ CO_2}{2.2}$$

THE FORM AND COMPOSITION OF THE BODY FLUIDS

The body of the healthy adult male contains 60 to 65 per cent water and about 15 per cent water-displacing fat, the remainder of the body weight being composed of the cellular and supporting tissue solids. The total body water varies from individual to individual depending upon his fat content. The fat-free body water is constant. Women, as a result of higher fat content, have an average body water content of about 50 to 55 per cent of body weight In the obese individual, whose fat content may be as high as 50 per cent, the body water may represent as little as 35 per cent of body weight. These variations in body water with fat content point to the need for considering body build in assessing clinical situations involving loss of body water. The acute loss of 10 per cent of body weight in an obese individual is of more serious portent than a similar percentage loss in a lean individual.

About 40 per cent of the body water (25 per cent of body weight) has electrolyte composition very similar to that of blood serum and may be regarded as the extracellular water Sixty per cent of the body water (35 per cent of body weight) has an electrolyte composition characterized by a high content of potassium and phosphate and composes the intracellular water. Table 1 indicates the ionic composition of extracellular fluid as found in plasma, and, by contrast, the approximate ionic composition of the water of muscle cells, the most important reservoir of intracellular fluid.

The maintenance of the ionic concentrations shown in table 1 is a complex and interlocking function of cellular metabolism, the vascular sys-

their pK. Generally the fixed anions are stronger acids than hemoglobin or bicarbonate and can displace cation from either of the two in the manner described for HCl above. Hemoglobin and bicarbonate are neatly balanced with regard to acidic strength and work closely together in their buffering action. Reduction of hemoglobin, as in anemia, cripples the buffering power of the blood. When excess fixed anion enters the body, as during ketosis, bicarbonate is the chief buffer and suffers reduction in concentration. This condition is called *metabolic acidosis*. Conversely, when excess cation enters the body bicarbonate is elevated and the condition is called *metabolic alkalosis*. When CO_2 accumulates in excess, as in respiratory obstruction, or is lost, as in hyperventilation, hemoglobin is the chief buffer The two conditions are termed *respiratory acidosis* or *alkalosis* respectively. If the buffers are able to absorb the acid or alkali excesses or deficits without change in pH the condition is spoken of as *compensated*. When pH changes the condition is spoken of as *uncompensated*. An excellent, clinically oriented, discussion of the buffer equilibria in blood is found in the paper of Singer and Hastings (12)

PATHWAYS OF WATER AND ELECTROLYTE METABOLISM

Through the Skin and Lungs

The water that is continually evaporated from the skin and lungs is an important portion of the water lost by the organism. Under the usual circumstances of life for the normal individual approximately 25 per cent of the body's daily heat production is dissipated in this way: each gram of water evaporated allowing for the dissipation of 0.58 calories. Above 30°C (85°F) active sweating begins and the percentage of calories lost by evaporation increases rapidly so that at 40°C (104°F) approximately 35 per cent of heat loss occurs in this fashion. Chilling of the body surface results in a decrease in the fraction of calories lost Approximately 40 per cent of evaporative water loss occurs through the respiratory tract During hyperpnea the fraction lost in this way increases and that through the skin decreases in a compensatory fashion so as to maintain thermal equilibrium

Since the basal heat production in normal persons at all ages is about 900 calories per square meter per 24 hours the basal evaporative water loss will be about 388 cc. per square meter per 24 hours $\left(\frac{900 \text{ cal.} \times 25\%}{0.58}\right)$ For the average 70 kg. man this will be about 10 cc per kg. per 24 hours. For an average infant of 7 kg., because of his greater surface area per unit of weight, the water loss will be about 20 cc. per kg. per 24 hours For the clinician, interested in evaporative water loss for the estimation of fluid requirements during disease, the basal loss provides, at best, only a working guide Increases in activity, body temperature, environmental tempera-

accounts for the difference in ionic patterns of the two phases. Death or injury tends to abolish the ionic gradients between the two phases so that potassium and phosphate are lost from the cells, and sodium and chloride tend to enter. During abnormal metabolic states such as dehydration, acidosis and alkalosis, dietary ion deficiencies, adrenal disease and diabetes similar partial rearrangements of ionic patterns take place. Ultimately the metabolic state of the intracellular fluid is the major determinant of the distribution of body water and salt.

REGULATION OF THE ACID-BASE BALANCE

Adjustment of the acid-base balance is maintained by the regulated excretion of carbon dioxide by the lungs and of cations and nonvolatile anions by the kidneys. Since, in terms of chemical reactions, the excretory processes are relatively slow, adjustment to sudden influxes of anion and cation to the body fluids are made by the buffer systems of the body fluids. The process may be likened to the water behind a dam: in the mass of water pH regulation is accomplished by buffering, while the excess anions constantly flowing in are spilled over the top as represented by the lungs and kidneys Chemically a buffer, when present in a solution, increases the amount of anion or cation that must be added to change the pH of the solution. A buffer consists of a weakly ionized acid and its strongly ionized salt acting as a pair A typical buffer pair are carbonic acid and its strongly ionized salt, sodium bicarbonate. When a strong acid, such as hydrochloric, is added to a solution of sodium bicarbonate sodium chloride and carbonic acid are produced. A neutral salt and a weak acid are produced in place of the original strong acid Much less pH change occurs than if the acid were added to a solution of sodium chloride. The neutral salts may be excreted by the kidney and the carbonic acid via the lungs. The relationship of the bicarbonate buffer pair to pH is expressed in the following relationship (the Henderson-Hasselbalch equation):

$$pH = pK + \text{Log} \frac{[NaHCO_3]}{[H_2CO_3]}$$

This simply states, stripped of its mathematical terminology, that the pH of a solution containing carbonic acid and sodium ions will be directly proportional to the ratio of the concentration of sodium bicarbonate to carbonic acid in the solution The chief buffer anions of the extracellular fluid are hemoglobin and bicarbonate These two anions have the property of changing their concentration (as anions) with changes in CO_2 or pH The "fixed anions", chloride, phosphate, lactate, etc , are so called because their respective concentrations are unaffected by pH changes in the physiological range. Buffer anions vary in their acidic strength, expressed as

position of diarrheal stools indicate that the sodium and chloride concentrations in stool water are about half those of extracellular fluid while the potassium concentration is from 2 to 10 times that of the extracellular fluid. Consequently diarrhea, in addition to producing deficits of water, sodium and chloride leads to excessive loss of the intracellular cation potassium. Potassium losses incurred during vomiting and suction drainage are insignificant when compared with those in diarrhea.

Accurate clinical assessment of gastrointestinal tract losses is difficult if not impossible without elaborate collecting procedures and provision for chemical measurements. Suction drainage losses may be measured accurately provided the quantities of fluid used to irrigate the tubes are taken into account. In infants stool losses as high as 10 cc. per kg. per 24 hours frequently are encountered. Sustained losses as high as this are rare in adults. Estimation of the chemical composition of such losses would seem relatively simple in view of the discussion just given. In practice this is not the case, the composition of the losses during vomiting, suction drainage and diarrhea is highly unpredictable and variable. This is necessarily so since the lost fluid is a mixture of varying amounts from different levels of the bowel. Volume for volume replacement of gastrointestinal losses with so-called physiological saline solution generally works well for short periods. When replacement is necessary over many days, however, edema generally develops, indicating that too much sodium is being given. In practice the fluids removed by suction drainage, like diarrheal stool, tend to be hypotonic with regard to total electrolyte concentration For this reason replacement with half concentrated physiological saline is less likely to lead to this complication.

Through the Kidney

While the skin and gastrointestinal tract losses of water and electrolytes are obligatory, those of the kidney are selective and regulatory. The renal excretion of water and electrolytes, within limits, is accommodated to the obligatory losses and the intake in a fashion that maintains constancy of volume and composition of the body fluids.

Water excretion is guided by the antidiuretic hormone (ADH) of the anterior pituitary, by the limitation imposed by the solute concentrating ability of the kidney and, probably, in some way by factors related to plasma or extracellular fluid volume The first two physiologic mechanisms are rather well defined and understood, the third is inferential and unproved "Osmoreceptors" exist in an area of the brain fed by the carotid artery. They have the ability to decrease or increase the rate of ADH production by the post-pituitary in response to changes in osmotic pressure of blood plasma. These osmotic reflexes seem mediated chiefly by the

ture and metabolic rate all increase this loss of water. In the average sized patient in a non-sweating situation the evaporative water loss will be about twice the basal level or 1200 cc. per 24 hours. Febrile patients living in hot humid environments may lose up to 5 to 6 liters a day. It is essential to bear in mind that skin and respiratory tract water loss is obligatory and continues unabated during water deficit, even to the detriment of the renal water requirement.

A constant loss of electrolyte through the skin occurs even though no sweating is occurring In the normal, non-sweating individual on a usual diet the daily loss of sodium, chloride and potassium is of the order of 2 to 4 meq per 24 hours. When active sweating occurs the losses are much greater and depend upon the intensity of the thermal stimulus, skin temperature and adrenal activity. In a general way, the greater the rate of sweating the higher is the concentration of sodium and chloride and the lower the potassium Hence, the net loss of sodium and chloride from the body increases greatly but the loss of potassium only moderately as the rate of sweating rises In the normal individual exposure to heat leads, over the course of 3 to 8 days, to a reduction of the concentration of sweat sodium and chloride at any given rate of sweating. As much as 150 meq of sodium (9 gm NaCl) may be lost in the sweat of an unacclimatized person undergoing moderate sweating Successful acclimatization will reduce the loss to a half or even a quarter of this value In hypoadrenocorticism the concentration of sodium and chloride in the sweat is high and adjustment to heat stress is poor. This indicates that adrenal activity is a *factor in controlling the loss of salt through the skin* The concentration of these ions in sweat is a useful aid in the assessment of adrenal cortical function (13, 14). Detailed descriptions of skin and respiratory tract water and salt losses under normal and extreme physiological conditions are found elsewhere (15, 16, 17) For those interested in industrial and military medicine the monograph of Adolph (18) is valuable.

Through the Gastrointestinal Tract

The fluid contents of the stomach and small bowel are considered to be in osmotic equilibrium with blood plasma Gastric juice contains relatively more *chloride* than sodium, duodenal fluid the reverse, while ileal fluid has a composition approximately that of extracellular fluid Loss of gastric juice leads to chloride deficit with alkalosis while loss of duodenal fluid leads to acidosis. Theoretically the loss of ileal fluid should lead to no disturbance in the composition of the body fluids other than simple diminution of the volume of extracellular fluid As ileal fluid passes through the large intestine its volume decreases and the concentrations of sodium and chloride fall while that of potassium rises Available analyses of the com-

by elaboration of a urine of maximal specific gravity (about 1.040). On normal intake of food this maximal concentration of the urine will be reached when about 1000 ml. of water are available for urine formation. If less than this quantity of water is available retention of solutes in the body will occur. When the concentrating ability of the kidney is completely lost urine volume, with a normal food intake, must be in the order of 5000 ml. per day if solute retention is not to occur. On diets containing no surplus solutes and sufficient non-protein calories (fat and carbohydrate) to spare protein catabolism maximally, urine volume, at maximal concentration, will be only about 200 ml. per day; if no concentration is possible, about 1000 ml. per day. These mechanisms provide maximal economy in the use of water for the excretion of urea and, at the same time, conserve or discard water in the interest of maintenance of the osmotic pressure of body fluids. When insufficient water is available at maximal urinary specific gravity, body water is sacrificed and solute retention occurs in a compromise effort to maintain homeostasis. Nitrogen retention is a usual accompaniment of dehydration. In renal disease, as is seen most clearly in advanced nephritis, the ability to adjust solute excretion to water intake is lost and a large volume of dilute urine is passed Modification of urine volume is possible only by changing the solute content of the diet. Consideration of these mechanisms constitutes one of the cornerstones in the physiological management of the patient with renal impairment, and is the basis for concentration and dilution tests of renal function.

Renal control of the excretion of sodium, chloride and bicarbonate determines the volume and composition of the extracellular fluid. According to present evidence, about 85 per cent of glomerular filtrate, having a composition similar to that of plasma, is reabsorbed in the proximal tubules with but minor changes in pattern or osmotic pressure In the distal tubules sodium, chloride and bicarbonate are reabsorbed by apparently independent processes that lead to maintenance of a constant composition of the extracellular fluid. One concept formulated as a working hypothesis to explain how such reabsorption is controlled is that of a balance between glomerular filtration and tubular reabsorption of the electrolytes (21). According to this concept, the distal tubes tend to reabsorb sodium in amounts that are constant If glomerular filtration supplies an amount to the distal tubules that exceeds the rate of reabsorption the ion will be excreted in the urine If a lesser amount is supplied by filtration all the filtered ion will be returned to the body. If, in the first instance, the rate of sodium excretion exceeds the rate of water excretion the plasma will become hypotonic and, through the ADH mechanism, water will be excreted until the osmotic pressure of the plasma is restored In the second instance the reverse sequence will occur Thus, the combined mechanisms could serve

concentration of sodium in the serum and are not invoked by changes produced by molecules freely diffusible into cells such as urea. The physiologic utility of such a device and its relation to the control of urine volume and concentration of urine and plasma is evident as one considers the following sequence of events: 1) Loss of water from the body occurs; 2) Blood plasma sodium is elevated as a result of concentration; 3) The osmoreceptors are stimulated and there is reflex production of ADH; 4) ADH leads to increased water reabsorption by the renal tubules, and 5) Osmotic pressure tends to be restored to normal by the dilution following increased reabsorption of water. Over-hydration of the body fluids leads to the same series of events but in reverse sequence. Blood plasma is filtered at the glomerulus at the rate of at least 180 liters a day, but of this volume, even under forced water diuresis or complete diabetes insipidus, only 10 to 15 per cent is ever excreted as urine. Eighty-five to 90 per cent undergoes obligatory reabsorption by the renal tubules. Within the residual 10 to 15 per cent range of water available for urine formation, variations in flow are controlled by ADH and by the need for excretion of the surplus salts of the intake and the solutes formed in the metabolism of protein (mainly urea) ADH secretion can be induced by emotion, exercise, syncope and by a number of chemical and hormonal agents. Since water equilibrium may be maintained at any volume of the extracellular fluid, control of volume must operate independently, probably in relation to sodium excretion

Primary disorders of the ADH mechanism are relatively rare in clinical practice Diabetes insipidus results when hypothalamic lesions block the reflex or as a result of destruction of the posterior pituitary. Rare instances of so-called "nephrogenic diabetes insipidus" have been encountered (19, 20), where ADH production is normal in response to the appropriate stimulus but the end-organ, the renal tubule, appears to be unresponsive Such cases must be carefully differentiated from those in which intrinsic renal disease has led to reduction of tubular function and consequent polyuria. The polyuria of advanced chronic nephritis is unmodified by the injection of posterior pituitary extracts.

Interlocked with ADH control of urine volume is the regulation imposed by the need to excrete the surplus solutes (salts) of the dietary intake plus those resulting from the catabolism of protein. When water is at a premium to the body, the volume of urine will be dictated by the total quantity of solutes requiring excretion and the maximal degree of urinary concentration that the kidney is capable of attaining When a surplus of water is available solute excretion will be independent of urine volume, and the ADH mechanism will act to control volume The normal kidney of man is capable of achieving a five fold concentration of glomerular filtrate

content of potassium is decreased and probably during acidosis The initial increased excretion of potassium that occurs when the salt-retaining hormone of the adrenal cortex is administered is probably related to the concomitant retention of sodium rather than to a specific renal effect. Any process that leads to retention of sodium with a tendency to elevation of extracellular fluid *concentration* of this ion leads to *increased* urinary excretion of potassium until equilibrium between intake and output of sodium is reached Abrupt increase in sodium intake leads to urinary loss of potassium. Conversely, loss of sodium leads to *retention* of potassium. If the intake of potassium is abruptly increased a reciprocal loss of sodium occurs until urinary excretion of potassium matches the increased intake.

When renal function is greatly impaired, as in advanced nephritis, an important difference between the excretion of the two ions becomes apparent. Since sodium is present in high concentration in glomerular filtrate, the quantity filtered always far exceeds the intake, even when filtration is greatly reduced If tubular function is reduced reabsorption of this large quantity of sodium fails *so that the patient continually faces the hazard of rapid sodium depletion* The converse holds for the renal excretion of potassium Since this ion is carried in the plasma at low concentration a relatively small amount is filtered daily. Reduction of filtering capacity below daily intake of potassium may lead to retention with its adverse cardiac effects As a result of these occurrences, the patient with advanced renal failure may resemble the patient with adrenal insufficiency

In addition to their excretory and osmoregulatory function the kidneys also act to control the acid-base equilibrium of the body by regulating the acidity of the urine. The excretion of a urine more acid than blood plasma may be accounted for by the reabsorption of alkaline buffer salts from glomerular filtrate by the tubules (23) *An alternate theory explains the* acidification by postulating exchange of hydrogen ions produced in the tubules for sodium and potassium of the glomerular filtrate (24) Whatever the ultimate mechanism may prove to be, it is capable of producing as much as 150 meq of free acid a day under the stress of severe metabolic acidosis Measurement of acid production may be accomplished by determining the quantity of base needed to adjust the urine to pH 7 4—that of blood plasma *The maximal urinary acidity is about pH 4 5* Accessory to the production of an acid urine in the excretion of the surplus anions formed during energy metabolism is the regulated excretion of the cation ammonium by the renal tubules Exchange of ammonium for sodium, potassium, calcium and magnesium of the glomerular filtrate allows for the excretion of large quantities of fixed anion without removal from the body of these essential components of body fluid structure An alkaline urine is *formed when the quantity of base requiring excretion exceeds that*

to control both volume and concentration of the body fluids Such a hypothesis, however, is difficult of proof. The rate of filtration and reabsorption of sodium is necessarily so great that barely measurable changes in filtration rate or concentration of sodium can lead to the excretion of very large amounts of this ion. Recent evidence indicates that changes in plasma volume, or some associated factor, play a role in the renal excretion of water and salt. This evidence is inferential and indicates that "volume receptors", analogous to those controlling osmotic pressure, exist in the cephalad portion of the body. Other factors that are known to act upon sodium reabsorption are the hormone of the adrenal cortex and changes in venous pressure

The processes by which the control of the renal excretion of potassium are achieved are just beginning to be studied Since the blood plasma perfusing the kidney contains potassium in relatively trace amounts, in comparison with its concentration in cells, the mechanisms that have been hypothesized to explain the control of excretion of extracellular electrolytes cannot apply to those which control the excretion of this important component of the intracellular osmotic pressure. Potassium is filtered at the glomerulus and selectively reabsorbed by the tubules. In addition, active secretion of potassium into the tubular urine occurs in man, since more of this ion can appear in the urine than can be accounted for by glomerular filtration. Probably the concentration of this ion in renal tubular cells acts, so to speak, as a pilot area that is able to initiate secretion or reabsorption of potassium into or from glomerular filtrate in accordance with the needs of the intracellular fluid as a whole.

Experimental evidence indicates that potassium secretion can be accounted for by ion exchange with the sodium of the tubular urine Mercurial compounds appear to block the tubular secretion of this ion. Increased excretion of this ion occurs, while intake is constant, when serum levels of potassium are high, during alkalosis, during periods when sodium retention is occurring and in response to mercurial diuretics if concomitant alkalosis is produced. Certain rare types of renal disease characterized by relatively normal glomerular function but with abnormal tubular function have been described in which renal excretion of potassium in excess of intake occurs It is characteristic of patients with such lesions that they tend to produce an alkaline urine and, as a result, develop acidosis An analogous situation can be produced in normal persons by the administration of sulfonamide compounds which inhibit the action of the enzyme carbonic anhydrase The theoretical implications of these observations in terms of the mechanisms by which potassium is secreted into the tubular urine have been discussed by Berliner and associates (22) Relatively decreased excretion of potassium occurs when serum levels of the ion are low, when the body

rapid respiratory rate should always raise the question of respiratory alkalosis and steps be taken to differentiate it from acidosis. The initial methods of treatment of the two are different and often crucial as to outcome as will be discussed below. Evidence of abnormal pigmentation of the skin and mucous membranes in the dehydrated patient should raise the question of adrenal insufficiency.

Progress in the study of body fluid physiology has brought forth a large number of laboratory examination of increasing complexity in performance and interpretation. Many of these are, because of the length of time required for their performance and their high cost in personnel and equipment, of little value for the clinician. The determination of glomerular filtration rate and renal blood flow by inulin and diodrast clearance, the measurement of total body water by isotope and antipyrine dilution and the determination of extracellular space and plasma volume are primarily research tools. In addition to their difficulty of determination, controversy still exists as to the meaning or validity of many of these determinations.

The determinations of hemoglobin or plasma protein concentration or hematocrit serve as an index of the degree of hemoconcentration, and hence, indirectly as a measure of plasma volume and extracellular fluid volume. Up to a certain point plasma volume is maintained at the expense of the whole extracellular fluid during dehydration. Beyond this point the plasma volume diminishes with concurrent rise in the non-diffusible constituents of the blood. Elevation of hemoglobin or plasma protein concentration, therefore, indicates advanced loss of extracellular fluid. Loss of blood, or plasma protein depletion will tend to vitiate the clinical value of these determinations.

The blood non protein nitrogen (N.P.N.) or urea serves as an index of the efficiency of renal function. Here again, moderate degrees of impairment, either functional or anatomic, do not lead to elevated values. Elevation of N.P.N. concentration indicates serious inability of the kidney to deal with the required solute load. Azotemia, aside from that resulting from anatomic destruction of the kidney, occurs when blood flow to the kidney is reduced as a result of decreased plasma and extracellular fluid volume during dehydration. N.P.N. concentrations above 75 mg. per cent are rarely a result of this type of disorder alone. Rehydration of the patient leads to a rapid decrease to normal concentration if renal disease is absent. The elevation of N.P.N. seen in congestive heart failure has a similar genesis.

Determination of the serum CO_2 content of blood drawn under oil furnishes a measure of the cation available to form the buffer anion bicarbonate—the alkali reserve. This procedure is technically simpler and preferable to the older determination of CO_2 combining power. Accessory to

of the fixed anions such as chloride and phosphate. This is accomplished by non-reabsorption of bicarbonate from the tubular urine. With vomiting, chloride is lost from the body in excess of base and the blood plasma becomes alkaline. The immediate renal response to this occurrence is the excretion of an alkaline urine. However, as base depletion of the body fluids progresses, the urine becomes acid even though systemic alkalosis persists. The attempt at preservation of the acid-base balance seems to be sacrificed in favor of support of the osmotic pressure.

The mechanisms carrying out the renal regulation of acid-base balance, like the excretory processes, require an adequate circulation of blood as well as water and electrolytes. The initial aim in therapy of the dehydrated acidotic patient is toward restoring these deficiencies. In renal disease the regulation progressively fails and urine pH and tonicity approach that of blood plasma.

DISTURBANCES IN THE BODY FLUIDS DURING DISEASE

Clinical and Laboratory Examination of the Patient

The clinical examination of the patient should include an effort to ascertain the length of time the disturbance has been present. Dehydration of relatively short duration will most likely involve predominantly extracellular deficits of electrolyte. When dehydration has been progressive for many days due to vomiting or diarrhea, loss of intracellular electrolyte is certain to have occurred and provision for its parenteral return may be necessary If possible, the weight prior to illness should be ascertained. The degree of water loss or dehydration may often be roughly estimated from such knowledge A history of renal disease should always be sought Reduction in renal function complicates the maintenance of water and electrolyte balance. The use or potassium in therapy is contraindicated in patients with renal disease until estimates of the degree of loss of function are available Evidence of anuria or oliguria should be searched for. Anuria is indication for specialized laboratory examination and therapeutic restraint until the degree of distortion of the body fluids is established Physical examination should include, if at all possible, accurate measurement of the body weight Changes in weight are the most accurate index of changes in body water Body build should also be assessed Equal losses of body water are of more significance in the obese than in the lean individual Cardiac function, with particular regard to the peripheral circulation and the presence of shock should be evaluated Skin turgor and the degree of moisture of the mucous membrane are a guide to the degree of dehydration. The character of the respiration is a guide to the state of the acid-base balance. Deep, labored and moderately rapid respiration may indicate acidosis as well as cardiac or pulmonary disease. An extremely

intracellular fluid, thereby delaying reduction of the former below the critical level at which circulatory impairment begins. This transaction requires loss of body potassium. Increased activity of the adrenal cortex, possibly mediated through changes in plasma volume or some associated factor, *must play a defensive role in dehydration. Such hormonal activity* would tend to limit sodium excretion and facilitate the excretion of potassium. According to experimental evidence about equal volumes of water *are lost, along with their quota of electrolytes, from both fluid compartments.* As long as the compensatory processes do not fail the body fluids retain their proper osmotic pressure. Thus, a severely dehydrated patient can be assumed to have lost about 100 ml. per kg. of water and about 7 5 meq. per kg of both sodium and potassium.

When about 5 per cent of body water has been lost in such a process the only detectable physiologic impairment is manifested by a rise in pulse rate and subjective fatigue When 10 to 15 per cent of body water is lost clinical dehydration is evident and defense of plasma volume begins to fail with its subsequent hemoconcentration and circulatory failure When the loss exceeds 20 per cent, recovery, even with vigorous therapy, is doubtful Disease in any of the organs concerned with these compensatory mechanisms sharply limits survival during dehydration Thus, in renal disease where the concentrating and salt-retaining powers of the kidney are lost, in adrenal insufficiency or posterior-pituitary disease water loss or deprivation quickly leads to serious difficulty It should be borne in mind that pure water deficit is a rarity in clinical medicine. Loss of extracellular and intracellular electrolyte through vomiting, diarrhea or sweating, by deficient renal control of electrolyte excretion or through adrenal insufficiency generally complicates clinical dehydration. The loss of electrolyte accentuates water loss and vice versa Definitive repair of dehydration can never be accomplished by water or salt alone

While total body deficit of water and salt generally accompanies dehydration it should be borne in mind that expansion of a part of the body fluids may leave the whole body with deficient water and salt to provide the proper quota for the heart and kidneys Trauma, burns, inflammatory reactions, and extensive frostbite may lead to functional deficits even though total body water and salt are normal or increased

Edema

Edema is generally considered to be primarily an increase in the volume of the interstitial fluid Whether the intracellular fluid and plasma share in the increased volume or are decreased in volume is a subject of controversy at the present time The mechanism of edema formation at the tissue level is adequately explained by the Starling principle relating capillary

the measurement of the CO_2 content is that of the serum chloride concentration. Under most circumstances the sum of these two measurements, in meq /l., remains equal to the sum of the normal concentrations. As a *first approximation, the sum of the two plus 15 may be taken as a rough* measure of the serum sodium concentration. Some of the exceptions to these rules of thumb will be noted below Serum pH is rarely measured in clinical practice. This is unfortunate as it represents the truest measure of the state of the acid-base balance of the body. Rapid, reliable and economical methods for this determination are available without resort to expensive electrode techniques (25)

The determination of the serum sodium concentration is primarily useful as an index *of the tonicity of the body fluids as 90 per cent of the extra*cellular fluid total base is accounted for by this ion Comparison of sodium concentration with the sum of chloride and total CO_2 plus 15 will give a relatively reliable measure of the degree of fixed acid accumulation, as a study of Table I will indicate The determination of serum potassium concentration serves as a rough index of the state of the intracellular stores of this ion.

Examination of the urine should include qualitative tests for albumin, glucose and acetone Testing of urine pH with nitrazine indicator papers often will identify the early stages of alkalosis. Measurement of urinary specific gravity during water deprivation is still an excellent test of renal function. Specific gravity may also be used as a guide to the degree of dehydration, particularly during parenteral therapy of the patient with normal renal function. The measurement of the electrolytes of the urine as a guide to therapy has a certain usefulness, especially in identifying marked salt depletion The use of urinary chloride concentration in following the therapy of the post-operative surgical patient has been described in detail by Scribner (26) Urinary chloride is often taken as an index of sodium excretion. This assumption is misleading as differential excretion of the two ions occurs under many conditions, particularly when low sodium diets are employed.

Dehydration

In the normal individual the loss of water alone may be considered to invoke the following ideal train of events 1) Decrease in extracellular fluid volume with a tendency *to concentration of its solutes, 2) as a consequence* of increased tonicity, water with its attendant potassium is transferred from the intracellular to the extracellular space and, simultaneously, 3) the ADH mechanism is set in action to limit water excretion, 4) the transferred potassium is excreted This sequence allows for sharing of the water deficit between the extracellular fluid and the large reservoir of

Sodium restriction, intravenous albumin therapy, diuretics, digitalization in congestive failure and adreno-cortical hormone therapy are the chief therapeutic agents used to lessen the tendency to edema or to initiate diuresis in edematous states. Reduction of sodium intake is the most effective method for decreasing the tendency to edema Extremely rigid sodium restriction, less than a gram a day, is often necessary to initiate loss of weight in congestive failure. Such low sodium intakes are not without danger as skin and gastrointestinal tract losses may lead to salt depletion. Reduction of salt intake to 1 to 2 grams a day is usually sufficient to control the progression of the edema and lessen the danger of salt depletion. Reduction of salt intake reduces the need for water restriction. The optimal fluid intake, in the absence of high environmental temperatures, is between 2000 and 3000 ml per day. The use of extremely high water intakes (as high as 4000 ml. per day) in conjunction with sodium restriction and acid-ash diets has not received wide acceptance. If such a regimen is followed water intoxication should be kept in mind as a possible consequence A recent adjunct to sodium restriction is the use of ion exchange resins. These compounds are insoluble, non-absorbable polymers which are able to exchange their hydrogen or ammonium ions for sodium, potassium, calcium and magnesium, in a solution Resins are now available that have a relatively high affinity for sodium and low tendency to exchange potassium When fed by mouth they carry with them into the feces a substantial portion of the ingested sodium but less of the potassium, thus lessening the tendency to potassium depletion Sodium restriction is still necessary but to a lesser degree Diets containing 2 to 4 grams of salt are feasible when such compounds are used and the palatability of the diet greatly increased. Hydrogen and ammonium exchange resins should not be used in the presence of renal failure because of their tendency to produce acidosis As clinical experience with sodium restriction has increased it has become evident that sodium depletion is an ever present danger Patients with sodium depletion present evidence of rapid dehydration, shock, oliguria or anuria, azotemia and muscular cramps. The condition may follow episodes of sweating, mercurial diuresis and the mechanical removal of edema fluid and ascites. The problem of diagnosis and therapy will be considered below

The use of plasma or concentrated salt poor albumin has had extensive trial in the management of the edema of the nephrotic state In general the results have been disappointing In most instances the injected albumin or plasma is almost quantitatively excreted in the urine in a relatively short period of time Furthermore, suggestive evidence has accumulated that prolonged use of such substances may lead to further reduction of renal function However, the use of albumin, in conjunction with paracentesis, will often result in diuresis in the nephrotic child Such combined

hydrostatic pressure to the opposing colloid osmotic pressure of the plasma proteins. If hydrostatic pressure is elevated or the plasma protein concentration decreased transudation of water and its attendant electrolyte will occur and edema result. The operation of this principle as stated is complicated by variable tissue tension and variation in lymphatic drainage. Application of this principle as an explanation of generalized edema, particularly to the maintenance or removal of excessive fluid, is not always apparent. The edema of the nephrotic state may increase or decrease without significant change in plasma protein concentration, or capillary pressure. *The formation of edema in terms of the Starling principle demands, as a corollary, renal retention of sodium and water.* Any factor which tends to modify the renal excretion of sodium will complicate the interpretation of the formation and removal of edema in terms of this principle Simple transudation of a plasma ultrafiltrate would quickly exhaust the plasma volume and arrest the process unless the supply of material is maintained. The mechanism by which the supply is maintained by the kidney in such an abnormal fashion is a source of controversy and much research Two theories are currently held to explain the edema of cardiac failure According to the "backward-failure" theory the following cycle of events causes edema: 1) elevated pulmonary and systemic venous pressure occurs as the primary event, 2) capillary pressure is elevated, 3) transudation of an ultrafiltrate occurs, 4) the plasma volume decreases and 5) a deficient cardiac output results. The fall in plasma volume or rise in venous pressure accounts, in the backward failure theory, for the renal retention of sodium and water How these stimuli operate is uncertain and, at the present time, largely inferential In the "forward-failure" theory the following sequence is said to occur: 1) a deficient output initiates events, 2) renal ischemia with reduction in glomerular filtration rate (as described previously) leads to sodium and water retention, 3) plasma volume and capillary pressure are increased and, 4) edema and increased venous pressure follow This theory has been questioned on the basis of the methods used to measure the increased plasma volume required of the theory Similarly, a decreased filtration rate does not always accompany congestive failure nor does filtration rate, if low, always rise when diuresis occurs Detailed critical discussions of these points of view and of the problem of edema in general are available (27, 28) Similar controversies exist concerning the mechanisms of edema formation in the nephrotic state, cirrhosis of the liver and toxemia of pregnancy Whatever the precise se-

edema; protein deficiency, reduction in blood volume, reduced plasma sodium concentration, reduced glomerular filtration rate, increased venous pressure, increased adrenal activity and ADH production.

normal sodium concentration accentuates the failure. On the other hand, certain patients with the "low salt syndrome" are benefitted by such therapy. Differential excretion of sodium and chloride is common in patients with congestive failure in conditions other than mercurial diuresis. The use of diets low in sodium but high in chloride, the usual composition of low salt diets, the administration of ammonium chloride and the differences between extracellular-intracellular exchanges all contribute to differential excretion and prejudice accurate assessment of electrolyte balance by inference from urinary and serum chloride measurements. The use of acidifying salts and mercurial diuretics in the management of the edema of the nephrotic state is hazardous because their effects depend upon adequate renal function. Severe degrees of acidosis and further renal impairment are frequent sequels to such attempts.

Changes in Tonicity of the Body Fluids

Hypotonicity (hyponatremia) of the body fluids occurs when sodium is lost in excess of water or when the intake of water exceeds the possible rate of excretion. In many instances the reason for the discrepancy is evident in the clinical history while in others the cause is obscure. Excessive sweating accompanied by a high intake of water, the administration of glucose solutions to the salt depleted patient and injudicious attempts to induce diuresis during anuria by water administration are all obvious causes of hypotonicity. The condition is also seen at times in diabetic acidosis and is a cardinal feature of adrenal insufficiency. Some degree of hyponatremia is a constant occurrence in congestive heart failure, the nephrotic state and the edema occurring in advanced cirrhosis of the liver Aggravation of the low sodium concentration to the point of production of symptoms may occur during therapeutic procedures designed to remove edema Hypotonicity of a fatal degree may occur during the diuresis following ACTH therapy of the nephrotic state. The mechanism leading to reduction of sodium concentration during various types of diuresis is obscure Suggestive evidence indicates that disturbances in the ionic structure of intracellular fluid may be the initiating event Hypotonicity is accompanied by shift of water into the intracellular fluid and by loss of water and salt from the vascular system. These two occurrences probably account for the physiological disturbances seen When the concentration of serum sodium falls much below 130 meq per liter urine volume begins to decrease and azotemia to appear, peripheral circulatory collapse follows, accompanied by profound weakness and muscular cramps If the decline of sodium concentration is produced rapidly convulsions may occur The diagnosis is made on the basis of these symptoms and signs and the finding of a decreased serum sodium or total base concentration In many instances of

procedures should be reserved for periods in which edema and effusion have reached the point of being detrimental to respiratory and cardiac function. The diuretic effect of albumin is probably dependent upon expansion of plasma volume with resulting increased glomerular filtration rather than upon elevation of plasma protein concentration. The use of solutions of acacia to induce diuresis is mentioned only to be condemned because of its tendency to accumulate in the liver and spleen The effects of the newer "plasma extenders" like dextran and polyvinylpyrrolidine are untried in the therapy of edema Plasma and albumin therapy are contraindicated in the edema of congestive heart failure.

Acidifying salts, particularly ammonium chloride, have had extensive use in the management of cardiac edema Its action is accounted for by the acidosis. The ammonium ion is removed by the liver leaving the strongly acidic anion which must be excreted with a quota of fixed cation and water. If the kidney is able to produce ammonia the acidifying and diuretic effects will decrease when this base-saving mechanism rises to full activity in three to four days. Potassium chloride is also a potent diuretic and depends for its action upon the imperative need for its removal when present in excess in the body fluids Since the renal excretion of potassium is impaired *in congestive failure its use as a diuretic in such conditions is often followed* by partial failure of excretion and the production of toxic effects (29). Acidifying salts have the ability to potentiate the action of mercurial diuretics. The diuresis obtained with mercurials leads, in the majority of instances, to substantial alteration in the pattern of serum electrolytes As the diuresis proceeds a greater quantity of chloride is excreted than sodium. The need for electroneutrality in the urine is met by increased excretion of potassium, ammonium and titratable acidity. The end result of this [illegible]

appearance of unresponsiveness. The administration of this salt to the unresponsive patient restores responsiveness and, if potassium intake is normal, leads to retention of this ion and restoration of serum potassium concentration (30) The common error of assuming that the serum chloride concentration may be used as an index of sodium concentration is well illustrated in this situation In the majority of such instances of hypochloremia the serum sodium concentration is normal or only slightly re- [illegible]

Determination of sodium concentration, pH of both is [illegible] management and diagnosis. The administration of hypertonic sodium chloride solution to patients with congestive failure with hypochloremia but a

indicated more by physiological status than by the finding of chemical abnormality.

Acidosis and Alkalosis

Metabolic acidosis occurs when there is a deficit of cation available to form serum bicarbonate. This may be brought about in two ways, either by relative increase in fixed anions or by relative loss of cations. As the more strongly acidic fixed anions enter the extracellular fluid they react with bicarbonate to produce CO_2 and the salt of the acid. Removal of this newly formed CO_2 accounts for the typical breathing pattern of acidosis. The administration of acidifying salts and the endogenous production of fixed acids during diabetic acidosis, methyl alcohol intoxication, starvation, shock, hemorrhage and exercise all may lead to accumulation of fixed acids in excess of the kidney's ability to excrete them. In advanced renal failure, phosphate and sulfate accumulate to a degree sufficient to lead to bicarbonate reduction. Relative cation deficit occurs when the renal mechanisms for ammonia formation and acidification of the urine fail or when alkaline digestive contents are lost from the body.

The normal kidney responds to acid excess by producing urine of maximal acidity and, after two or three days, of maximal ammonia content. When the capacity of these base saving mechanisms is exceeded, the fixed bases, sodium, potassium, magnesium and calcium are excreted along with water. Dehydration becomes a secondary event as intracellular and extracellular cation are swept out. The patient with manifest acidosis always presents deficit of body water as well as of intra- and extracellular electrolyte.

Shifts of cation between extra- and intracellular fluid can account for acidosis. The body fluid compartments achieve considerable independence in regulation of acid-base balance by virtue of the limited exchange of their respective anions. Normally intracellular sodium is variable and can be transferred back and forth between compartments to a certain degree. Such transfers take place without equivalent exchanges of phosphate or chloride. Thus, movement of sodium into cells diminishes the base available to form extracellular bicarbonate and results in acidosis. Such events occur clinically and result in acidosis without change in total body electrolyte. Conversely, intracellular sodium can function to a certain degree in maintaining extracellular bicarbonate (31). The exchanges of cation between the body fluid compartments are an important consideration in the therapy of acidosis. The administration of alkali as sodium bicarbonate or lactate restores the extracellular fluid bicarbonate to normal. Simultaneously, however, the intracellular deficit of potassium is, to a certain extent, replaced by sodium. Thus, the administration of alkali will repair the acidosis

this condition the administration of salt effects dramatic improvement. If the hypotonicity is accompanied by dehydration the intravenous administration of normal saline is the treatment of choice. When edema is present or the body fluid volume is normal 5 per cent salt solution is indicated. Sufficient hypertonic salt solution should be given to elevate the cation concentration in all body water. 0.5 to 1.0 ml of 5 per cent salt solution per kg. of body weight per meq. lowering of the serum sodium concentration is usually required to return the concentration to normal. Rigid water restriction must follow such a procedure if it is to be effective. In successful cases diuresis begins within a few hours and the circulation improves. Such a procedure will work well in instances of salt depletion resulting from improper treatment with glucose solutions. However, in severe congestive failure it is often ineffective. No diuresis is produced even though the concentration of sodium in the serum is increased. Intense thirst follows in this instance, water restriction becomes difficult to enforce, the edema increases and the serum sodium concentration falls to its initial level.

So-called asymptomatic hyponatremia often is uncovered as a result of incidental determination of serum sodium or chloride concentration. This is seen at times in patients with advanced pulmonary and meningeal tuberculosis or with lesions of the central nervous system such as tumor and hydrocephalus. No signs of dehydration or shock are evident. *The administration of sodium chloride is without benefit. It is usually excreted quantitatively* without significant elevation of the serum concentration.

The reverse condition, hypertonicity of the body fluids, occurs when water depletion exceeds the loss of salt. It is always accompanied by dehydration and is a condition seen more often in children than in adults. Hypertonicity may follow the administration of excessive salt solution, the dehydration of diarrhea where large volumes of hypotonic stools are lost, during adrenal hormone therapy, during water restriction in patients with diabetes insipidus and in heat exposure when sweating is deficient. Hypertonicity has been described following certain types of sulfonamide anuria. Patients with hypertonicity may show symptoms of shock but evidence of circulatory deficiency is less prominent than in salt depletion. Cerebral symptoms, unconsciousness, convulsions, paralyses and hyperthermia are common. Treatment is directed toward restoring the deficit of water by the use of glucose solutions. If the hypertonicity is chronic, good physiologic adjustment may have been made as evidenced by relative lack of symptoms. In such instances, vigorous attempts to reduce the high sodium rapidly should not be made. Symptoms of salt depletion may develop while the serum sodium concentration is still considerably elevated above the normal. In general, *treatment of disturbances of tonicity is*

pressure of the two compartments are equalized in the presence of deficits in either area. Prolonged metabolic alkalosis reduces glomerular filtration rate, renal plasma flow and tubular excretory capacity with slow reversal when the alkalosis is relieved (33).

Treatment of metabolic alkalosis involves removal of the cause and, like that of acidosis, repair of the deficits of water and electrolyte that usually accompany the condition as seen, for example, in pyloric obstruction. If renal function is relatively normal the administration of isotonic sodium chloride and glucose solutions to the alkalotic patient usually adequately restores electrolyte balance On theoretical grounds the administration of potassium is indicated in such conditions. Study of electrolyte balances in patients recovering from vomiting likewise indicate the need for this ion. The actual clinical need for potassium must probably be judged on the duration of alkalosis and a consideration of how quickly oral feeding can be established. Infants with pyloric stenosis are usually seen early before large deficits of potassium have developed. Therapy with saline and glucose solutions adequately and quickly prepares them for operation and successful early feeding can be anticipated. In such cases potassium is not essential. In severe instances pre-operative preparation may require one to two days and parenteral potassium therapy should be employed. Chronic alkalosis with hypokalemia is often seen post-operatively in patients who have undergone severe surgical procedures and have required suction drainage and parenteral fluids The alkalosis and hypokalemia in such instances respond to potassium administration (34). The use of ammonium chloride and hydrochloric acid has been described as a method of treatment for metabolic alkalosis (35, 36, 37). The parenteral use of such acidifying substances should always be accompanied or followed by sodium chloride, potassium chloride and water if dehydration is present The administration of 4 to 5 grams of ammonium chloride a day to the alkalotic patient unresponsive to mercurials will restore serum concentrations to normal, restore responsiveness and lead to a positive balance of potassium if its intake is normal

Clinically the patient with this type of metabolic defect presents evidence of tetany, lethargy, weakness and a depressed respiratory rate How much of this is referable to the disorder of potassium metabolism and how much to the alkalosis is difficult to state The administration of ammonium chloride will abolish the symptoms and signs temporarily, but the condition tends to reoccur if potassium is not made available

Respiratory alkalosis results whenever, for any reason, respiratory activity exceeds that needed to maintain the equilibrium between CO_2 production and removal The primary feature of this type of disturbance is depression of the CO_2 tension Such a decrease may be found in hyper-

but will exchange one abnormality of the intracellular fluid for another and even accentuate the deficit of potassium Evidence is accumulating that a high intracellular sodium concentration may be at least as unfavorable as acidosis. The adverse symptoms of acidosis are as much referable to deficit of body water and electrolyte as to the acidosis *per se*. Relatively severe degrees of acidosis are well tolerated if the water and electrolyte content of the body are normal. Correction of acidosis without correction of dehydration usually accentuates the clinical condition. Restoration of the deficits of water and electrolyte allows for the correction of the acidosis by metabolic and renal activity *In severe acidosis repair of dehydration* with saline-lactate solution may be followed by alkalosis associated with hypokalemia. These chemical findings may be accompanied by gross abnormalities of the electrocardiogram, and muscular and respiratory paralysis. The administration of potassium corrects the chemical and clinical abnormalities. *Provision for parenteral potassium administration in the* therapy of acidosis should be made if oral feeding with high potassium containing foods like milk and fruit juice cannot be initiated within twelve to eighteen hours.

Respiratory acidosis results from the primary accumulation of CO_2 when pulmonary ventilation is interefered with. Narcosis, paralysis of the muscles of respiration, emphysema and bronchiolar obstruction all lead to the development of a high serum CO_2 content and, if severe enough, lowering of blood pH The serum chloride concentration tends to be slightly low and that of sodium high, although these abnormalities are never marked No significant changes in the distribution of body electrolyte are known to occur Treatment should be directed toward the pulmonary disease if possible Alkali therapy is contraindicated in this type of acidosis.

Metabolic alkalosis occurs when the fixed anion chloride is lost in excess of sodium or when alkaline salts are ingested Loss of chloride in excess of sodium occurs in vomiting, during high level intestinal suction drainage and during intensive mercurial diuresis As Darrow has shown, metabolic alkalosis also occurs when relative deficit of intracellular potassium is present and, conversely, the production of alkalosis leads to relative deficit of potassium (32). The administration of cortisone or ACTH leads to alkalosis and potassium deficit, partiularly if the dietary sodium is high Elevation of barcarbonate and serum pH with hypokalemia is a feature of Cushing's syndrome. The reciprocal relationship between extracellular fluid alkalosis and the concentration of intracellular potassium appears to be maintained by renal regulation The kidneys will not excrete sodium to decrease the serum bicarbonate unless potassium is made available The biological significance of these relationships is unknown; they probably represent the means by which changes in acid-base balance and osmotic

tracellular water is in the order of 140 to 160 meq. per liter. Thus, a ratio of approximately 40:1 exists between intra- and extracellular fluid concentrations of potassium. This gradient is an unstable one and presumably is maintained by the oxidative energy of the cells. Any process, such as anoxia, that interferes with maintenance of this gradient leads to loss of this ion from the cells. According to Darrow (32), a high degree of correlation exists between the concentration of extracellular fluid bicarbonate and the concentration of intracellular sodium and potassium. When extracellular fluid bicarbonate is high (alkalosis) intracellular sodium is high and both intra- and extracellular concentrations of potassium tend to be low. In acidosis the reverse appears to hold true. Darrow considers these relations to represent a biological equilibrium which is maintained when renal regulation of the body fluids faces a deficit of sodium, potassium, or chloride. The relation is termed "biological" in order to indicate that attainment of the equilibrium is a relatively slow process occurring in response to metabolic activity rather than the simple diffusion of ions. According to this view, deficit of chloride and deficit of potassium tend to produce the same changes in both the intra- and extracellular fluids. When equilibrium is reached in the presence of deficit of one of these ions a deficit of the other may be predicted In adrenal insufficiency the renal tubules fail to reabsorb filtered sodium and deficit of sodium and acidosis develops Concurrently the excretion of potassium decreases and a high concentration of this ion develops in the body fluids. In Cushing's disease or following the administration of desoxycorticosterone increased tubular reabsorption of sodium and increased excretion of potassium occurs. Consequent to this the concentration of potassium in the body fluids falls and the concentration of both intra- and extracellular sodium rises and alkalosis occurs Dietary deprivation of potassium may lead to a decreased concentration of intracellular potassium, increased cellular sodium concentration and alkalosis As has been indicated, dehydration leads to loss of potassium from the intracellular fluid Such losses represent, in large part, merely a contraction of the compartment It is only when the intracellular fluid volume is restored and potassium is not made available that the concentration of potassium is adversely affected. It should be emphasized that changes in intracellular potassium content are, in large part, conditioned by the intake of sodium at the time the changes are occurring Thus the production of potassium deficiency by dietary deprivation or adrenal cortical hormone administration occurs only when the intake of sodium is normal or high. Conversely the retention of potassium in adrenal insufficiency may be abolished by the administration of sodium. These relationships between the body fluid compartments are those found in experimental animals and in relatively pure and rare clinical states Clinical conditions involving

pyrexia, in lesions of the central nervous system involving the respiratory center, in encephalitis, in hysterical hyperventilation and in the apparently specific respiratory center stimulation occurring in salicylate intoxication. It may also result from the hyperventilation occurring in the anoxia of congestive heart failure and high altitudes. From the standpoint of acid-base balance, respiratory alkalosis may be regarded as an essentially inappropriate response. As CO_2 is removed by the increased respiratory activity, the blood pH increases and cation present as serum bicarbonate is transferred to hemoglobin. This process lowers serum bicarbonate and mitigates the degree of alkalosis—the normal ratio of carbonic acid to bicarbonate tends to be restored. This buffer effect is aided by the initial excretion of an alkaline urine which further tends to lower the serum bicarbonate concentration If, at this point, the stimulus to hyperventilation is removed true metabolic acidosis will be present. This sequence is seen in salicylate intoxication, the most commonly encountered type of respiratory alkalosis. Early in the course of the intoxication blood pH is elevated and the total CO_2 may be low. The finding of a low CO_2 coupled with the intense respiratory activity often leads to the mistaken diagnosis of metabolic acidosis. If alkali is administered at this point fatal tetany *may follow. Later in the course of the condition, as the effect of* the drug wears off, both blood pH and serum CO_2 are usually low and treatment of the acidosis with parenteral fluids may be necessary. The determination of blood pH is essential to the proper diagnosis and treatment of different stages of the intoxication. The use of narcotics in the attempt to depress the exaggerated respiratory activity of salicylism increases mortality in the experimental condition in the dog (38) Clinical experience in man substantiates these experimental observations

Disturbances of the Intracellular Fluids

While it has long been known that disturbances of the body fluids such as occur in diarrhea, diabetic acidosis and dehydration lead to abnormalities in the composition of intracellular fluid, clinical interest in these changes is of recent origin. This portion of the body fluids previously had been considered as inaccessible to replacement therapy In 1946, Darrow (32), basing his premise on extensive studies of the behavior of intracellular electrolyte in animals under various conditions and on balance studies in infants with dehydration, demonstrated the practicability of and rationale for the use of potassium in replacement solutions. In the intervening years much clinical investigation has substantiated his point of view and confirmed the therapeutic utility of potassium administration in certain disturbances in body fluids.

Analyses of tissues indicate that the concentration of potassium in in-

excreting less of the ion, but several days are required before maximal urinary conservation is achieved. If the intake of potassium exceeds the maximal rate of renal excretion, the ion will accumulate in the body fluids and eventually produce toxic effects. The maximal rate of excretion is conditioned by the magnitude of the previous intake. If this is gradually increased over the course of several days, the maximal rate of potassium excretion in the normal person can be greatly increased. This range of adjustment is decreased in patients with renal disease and with congestive heart failure. Gains or losses of potassium from the body must be measured against the simultaneous balance of nitrogen. The fasting individual loses *approximately 3 meq of potassium for each gram of nitrogen excreted.* This process represents wasting of body tissue and does not indicate deficit. It is only when the potassium to nitrogen ratio exceeds this value that deficit of potassium can be considered to be occurring

Patients with potassium deficiency exhibit certain rather non-specific symptoms and signs which, in association with a history of disturbances leading to a loss of this ion, suggest the possibility of the diagnosis A low serum potassium concentration and the characteristic electrocardiogram confirm the diagnosis. Muscular weakness and hypotonia are the earliest clinical signs. They may progress to frank peripheral and, later, respiratory muscle paralysis. Abdominal distension may be seen and is probably related to the atony of smooth muscle of the intestine. In experimental animals potassium deficiency produces paralytic ileus Signs related to the cardiovascular system may be found The heart is enlarged, murmurs appear, the pulse pressure rises and, finally, cardiac failure may occur The most frequently encountered clinical history that precedes such symptoms and signs is one of several days to weeks of reduced food intake associated with the *concurrent loss of body fluids and electrolytes through* diarrhea, suction drainage, vomiting and uncontrolled diabetes Surgical procedures punctuating such a train of events tend to increase the deficit The large quantities of sodium containing solutions that such patients usually receive to combat dehydration and acidosis play a large role in the development of symptomatic deficiency It is only when body fluid content is near normal and acidosis has been repaired that symptoms appear Prolonged treatment of peptic ulcer with alkali may lead to the potassium deficiency syndrome. In such instances the deficit is related to the high intake of sodium as well as the renal impairment that accompanies prolonged alkalosis (33). In Cushing's disease as well as during prolonged administration of adrenal hormones, low concentration of serum potassium with alkalosis appears. In such instances the alkalosis is refractory to the administration of sodium or ammonium chloride but does appear to respond to potassium chloride administration.

disturbance of the intracellular fluid are usually the result of multiple deficits of ions, often occurring in the presence of large loads of other ions and in the level of adrenal activity. These observations, however, provide the descriptive basis for consideration of the therapeutic approach to the intracellular fluid.

The potassium content of the human body is in the general order of magnitude of 50 meq per kg. of body weight. Thus, the body of the average adult contains about 3500 meq. of the ion. The extent to which this quantity can be reduced in man before symptoms appear is unknown, but is probably conditioned by the particular circumstances leading to and accompanying the deficit. Experimental animals maintained on low-potassium high-sodium diets lose about 25 per cent of their total body potassium when symptoms of deficiency are fully developed. Patients recovering from severe diarrhea and diabetic acidosis may retain quantities of potassium equal to about 20 per cent of expected normal content. Such patients may or may not exhibit laboratory or clinical evidence of deficiency. The validity of using retentions alone as an index of deficit tends to be weakened by the observation that normal human subjects may retain comparable quantities of potassium when their intake is suddenly increased (31). Patients, especially those with chronic illness, often have a low total body potassium content per unit of weight without demonstrable signs of potassium deficiency (39). In familial periodic paralysis all clinical and laboratory findings of potassium deficiency may develop without external loss of this ion. Patients with severe diabetic acidosis quite regularly exhibit elevated serum potassium concentrations and electrocardiographic signs of potassium intoxication despite the fact that large quantities of this ion have been lost from the body It is only when treatment of the acidosis with insulin and sodium containing solutions is instituted that evidence of potassium deficit appears The electrocardiographic abnormalities associated with potassium deficit may disappear before any appreciable repair of the deficit is accomplished. Conversely, the abnormalities may persist after the deficit is restored and serum concentrations are normal. The intracellular fluid seems, therefore, to act as a somewhat indifferent reservoir for potassium with rather wide limits for contraction or expansion. The appearance of clinical symptoms and signs must be related to factors other than concentration or total amount, possibly to factors associated with the state of the acid-base balance

The usual intake of potassium is widely variable. Infants on cow's milk feedings get 6 meq per kg per day while the average adult intake is 1 to 2 meq. per kg per day. After the demands for growth are met (1 to 2 meq. per kg. per day), the surplus is excreted, chiefly in the urine. If the intake of potassium is suddenly stopped, the kidneys respond by

sodium concentration if this is low and improving renal function. The administration of glucose and insulin will often transiently reduce serum concentration by retarding protein catabolism and prompting potassium uptake by cells as glycogen is formed. Hypertonic sodium chloride solution will restore sodium concentration and, at times, improve renal function This procedure, while theoretically useful, must be undertaken with restraint. Its beneficial effect depends, in part, upon the premise that the kidney will respond with a diuresis when the concentration and extracellular fluid volume are restored. If renal damage is so great that this cannot occur, fatal pulmonary edema may follow the attempt. The intravenous injection of calcium salts is useful but of transient effect. The artificial kidney and the administration of exchange resins by mouth or rectum have been used to promote extra-renal excretion of potassium.

PARENTERAL FLUID THERAPY

The aims of parenteral fluid therapy are fourfold. The first is the treatment of circulatory failure and resulting shock by restoration of the extracellular fluid and plasma volume The second is restoration of the body electrolyte and water. The third is provision for obligatory expenditure of water and electrolyte. The fourth purpose is provision for nutrition These aims are listed in order of clinical importance in the progression of therapy but they should be thought of, as treatment is planned and carried out, as simultaneously occurring needs. A corollary of these four aims is the treatment of the underlying disease where possible. Antibiotics, insulin and adrenal hormones, when specifically indicated, restore the fundamental metabolic defect that has led to the losses.

Successful and sustained parenteral fluid therapy cannot be effective unless renal function is reasonably competent For this reason the treatment of circulatory failure and shock by expansion of plasma and extracellular fluid volume is the initial step in repair of dehydration As the circulatory failure decreases, renal function improves and allows the ends of therapy to be accomplished. Isotonic sodium chloride solution, 5 per cent dextrose in saline, whole blood, plasma and albumin solutions are the fluids of choice for expanding the extracellular fluid In shock due to blood loss, whole blood and plasma remain the fluids most widely advocated. Where shock is primarily due to dehydration, saline solution alone or in combination with blood or plasma is indicated Fifteen to 30 ml. per kg of body weight given over a period of one to two hours usually restores the circulation to a point where renal function is sufficient to aid the subsequent therapeutic efforts Whole blood or plasma may be used in dehydration shock provided they are accompanied by the simultaneous administration of proper quantities of water and electrolyte. Five or 10 per cent glucose

The electrocardiographic changes associated with potassium deficiency have been described thoroughly. Lowered concentration of potassium, with normal calcium ion concentration, increases the tonicity of cardiac muscle. In the absence of potassium, the heart beat stops in the contracted state. During such a process, the electrocardiogram shows a lengthening of the Q-T interval, decreased height or inversion of the T wave and depression of the S-T segment. In the isolated heart, it has been shown that the rate of decrease of the concentration of potassium in the perfusate conditions the response If the concentration is slowly reduced, the electrical changes are minimal Conversely, rapid elevation of the concentration perfusing the heart conditioned to beat at low concentrations may lead to high potassium type of arrest at concentrations below the normal. These observations are of clinical value in that they help explain some of the discrepancies between serum concentrations and electrocardiographic findings often encountered. They stress the importance of avoiding rapid restoration of serum concentrations in hypokalemic individuals.

The margin of physiological safety with regard to potassium excretion is so great that retention rarely occurs It is only in the latest stages of renal failure or in complete anuria that potassium rises to toxic levels in the serum Acidosis, particularly that accompanied by low serum concentrations of sodium, is often associated with high concentrations of serum potassium This combination of changes is seen in adrenal insufficiency and in the "salt wasting" types of renal disease I have seen transient hyperkalemia with toxic manifestations appear in patients with leukemia treated with x-ray and anti-folic acid preparations Presumably, the rapid breakdown of cellular tissues liberates quantities of potassium in excess of the kidney's ability to excrete the ion Increased serum concentrations of potassium occur in diabetic acidosis because of the high cellular catabolism and the renal failure associated with the dehydration The clinical symptoms and signs that have been described in presumable association with hyperkalemia are mental confusion, paresthesias of the extremities, pallor, bradycardia and, occasionally, flaccid paralysis and cardiac arrest.

Increasing concentration of potassium in the serum reduces the contractility of the heart and eventually leads to arrest in the completely relaxed state. When serum potassium concentration rises much above 6.5 meq per liter, a definite series of events appears in the electrocardiogram. Initially, the T waves become peaked and elevated; secondly the QRS and P-R intervals progressively lengthen and auricular standstill may occur; thirdly is the appearance of a biphasic curve; and, finally, at about 10 meq. per liter of potassium, cardiac arrest.

The treatment of hyperkalemia is directed toward stopping the intake of the ion, attempting to reverse the metabolic transfer, restoration of serum

tance. The solution designed by Darrow (40) contains sodium and chloride in the ratio of extracellular fluid together with a quantity of potassium that may be administered safely if certain time limits for its administration are observed. Darrow's solution contains 4.0 gm. sodium chloride, 2.7 gm. of potassium chloride and 52 ml. of molar lactate per liter. If 50 to 80 ml. per kg. of this solution are given, the average deficit of extracellular water and electrolyte will be restored and 2.5 to 4 meq. per kg. of potassium will be administered. This quantity of solution must be given in no less than four hours and, preferably, in 8 to 12 hours or longer in order to avoid the toxic effect of potassium. This quantity of potassium will only partially repair the average intracellular deficit. Several days of such therapy are required for complete repair. The daily administration of greater quantities of this ion is potentially hazardous and probably exceeds the maximum daily rate of cellular uptake of potassium in ill patients. The intravenous or subcutaneous administration of potassium should not be initiated until a definite urinary output is established. Its use should be avoided if any clinical suspicion of adrenal insufficiency exists. Darrow's solution, as indicated, will restore the extracellular fluid deficit before the intracellular. Continued use of this solution, once the extracellular deficit is restored, will provide a surplus of sodium and chloride that many patients requiring prolonged parenteral maintenance will not tolerate. This solution is primarily designed for repair rather than maintenance. Potassium chloride may be administered in glucose solution or in hypotonic sodium chloride solutions for maintenance therapy as will be described subsequently.

Up to this point only the water and electrolyte required for restoration of the circulation and body deficit have been discussed. Provision must also be made for the losses of water and electrolyte that are occurring during the period of replacement therapy. These obligatory expenditures have been discussed under the sections on exchanges through the skin and lungs, gastrointestinal tract and kidneys. The average adult who is not sweating requires about 20 ml. per kg. per day for evaporative water loss. In the infant this requirement is approximately twice that of the adult. The water requirement for successful renal function may be taken as essentially equal to *insensible* water loss in both age groups. The water required to meet evaporative loss is furnished in parenteral therapy as glucose solution. Similarly, the renal water requirement, during the reparative stage at least, is best furnished as glucose solution. An understanding of the rationale for the use of glucose (water) solution *to cover these obligatory expenditures is* essential for intelligent therapy. If the total water requirement, i.e., for replacement plus obligatory loss, is given as isotonic sodium chloride solution, a surplus of electrolyte will remain in the body as evaporative water loss proceeds. If the body *fluids are not to become* hypertonic, this electro-

solutions should be avoided in the initial therapy of dehydration unless a very severe degree of hypertonicity of the body fluids is known to exist. Injection of solutions containing glucose alone is tantamount to the injection of distilled water as the glucose is quickly oxidized. An immediate favorable response may occur; only to be followed by return of the original clinical state as a consequence of the dilution (hypotonicity) of the extracellular electrolyte.

Once the circulation is relatively normal, attention should be focused simultaneously on the restoration of the deficits of water and electrolyte and upon the need for replacing obligatory expenditures. No sure formula can be given for such replacement. Reliance must be placed upon a knowledge of what the average deficits are and upon the ability of the kidney to conserve or reject the administered electrolyte and water in the proportions needed by the patient. The average severely dehydrated patient has lost 10 to 15 per cent of his weight as body fluid drawn about equally from the two body compartments. Repair of these average deficits would require 100 to 150 ml. of water. The extracellular loss, 50 to 75 ml. per kg., may be supplied with isotonic saline solution or, more precisely, with a mixture of one part M/6 sodium lactate or bicarbonate solution and two parts of isotonic saline. *The use of lactate-saline mixtures in the proportions mentioned supplies* sodium and chloride in about their ratio in extracellular fluid and plasma. The administration of lactate or bicarbonate solutions alone to restore the volume and acid-base equilibrium of the extracellular fluid is unnecessary, and at times, harmful as it accentuates the intracellular disturbance Balance studies repeatedly have shown that, when this is done, approximately half of the sodium administered is transferred to the intracellular fluid. Solutions composed of lactate and saline in a ratio of 1:2 serve adequately to repair acidosis and, at the same time, to expand the extracellular fluid The initial rapid infusion of 15 to 30 ml per kg used to restore circulatory competency serves to replace from a third to a half of the extracellular deficit. The remainder of the deficit (35 to 45 ml per kg) is best administered slowly over the first day of therapy.

In theory, the approximate half of the deficit of water lost from the intra- [illegible] restores the circulation and extracellular fluid deficit and leads to rapid recovery so that the ions and fluid necessary for intracellular repair may be drunk. Current advances in antibiotic and surgical therapy, however, have increased the number of patients who survive the initial period of shock and yet are unable to begin an oral intake In these, the need for parenteral therapy directed toward the intracellular fluid assumes increasing impor-

tice, however, the problem becomes increasingly complex and clinically difficult. Most patients requiring parenteral therapy beyond a period of 3 to 4 days are those presenting such severe clinical problems as intestinal obstruction with accompanying peritoneal infection, severe degrees of diarrhea, ulcerative colitis and frank renal or hepatic disease. In such critically ill patients, salt and water homeostasis becomes progressively more unstable. Often the desired urine volume and specific gravity fail to be maintained despite theoretically proper management. The kidneys may excrete excessive quantities of salt or fail entirely in this task. Hypotonicity may develop and be refractory to the administration of hypertonic saline solution. Dehydration may occur despite the apparently correct administration of water and electrolyte. Attempts to repair such dehydration often are followed by the sudden appearance of pulmonary or generalized edema. Such patients generally show evidence of progressive renal failure as manifested by rising blood non-protein nitrogen concentrations.

This failure of many patients to do well despite carefully carried out fluid and electrolyte administration has focussed attention on the fourth purpose of parenteral fluid therapy—nutrition. This apparent inability to maintain salt and water homeostasis may be attributed to nutritional failure, particularly with regard to nitrogen balance. A truly positive and sustained positive balance with a quantitatively and qualitatively adequate intake of nitrogen cannot be achieved by the organism unless the full caloric requirement is met. Protein loss can, however, be minimized by sub-optimal caloric intakes Gamble has shown that approximately 100 gm. of glucose per day for the average adult exerts maximal protein sparing effect. Increasing the quantity exerts no further effect until the full caloric intake is given Similarly, increasing the intake of protein or amino acids on subcaloric intakes exerts no positive effect upon the nitrogen loss (16). With these observations in mind the clinician faces two alternatives in the management of the patient dependent upon a parenteral intake for nutrition Sufficient glucose may be given to provide the maximal protein sparing quantity or an attempt made to provide a full intake of both calories and protein by the intravenous route. Amino acids and protein hydrolysates may supply the protein needs when combined with glucose. About 1 gm. per kg. per day is required by adults and 2 5 to 3 0 gm per kg for infants To meet the adult requirement for full calories about 3 liters of 10 per cent glucose or 2 liters of 15 per cent glucose will be required Positive balances of nitrogen in experimental animals and normal men may be achieved by such means Whether this end can be accomplished in patients with infection, fever, malignancy or severe trauma is controversial. The majority of evidence indicates that it cannot. A major difficulty encountered in attempts to provide a complete caloric and nitrogen intake by parenteral means is the

lyte must be excreted. In dehydration the kidneys already face the task of removing the urea and salts formed during metabolism in a minimal quantity of water. If only isotonic saline is given to cover the obligatory loss, the remaining sodium and chloride must compete with other metabolites for available renal water, the ability of the kidneys to concentrate solutes will be exceeded and retention of both metabolites and salt will occur. In actual effect the dehydration will be accentuated.

Thus, for the average severely dehydrated patient the quantities and types of fluid required to repair dehydration and simultaneously replace obligatory losses (assuming no gastrointestinal loss) would be approximately as follows: For the repair of tissue deficit, 50 ml. per kg. per day of isotonic electrolyte solution (saline, 1:2 solution, Darrow's solution), 20 ml per kg. per day of 5 to 10 per cent glucose solution for evaporative water loss and 20 ml per kg. per day of glucose solution for the renal requirement, a total of 90 ml. per kg. per day of fluid. For the infant and small child, because of his greater metabolic expenditure per unit of mass, usually approximately twice these quantities of fluid are given. These values are average ones based on severe deficits Less fluid is necessary when the dehydration has been milder. When the status of the cardiovascular system is in question, repair should proceed at a slower rate so that less total fluid per day is administered If the dehydration is hypotonic with regard to sodium concentration, as in adrenal insufficiency, larger quantities of electrolyte in the same total volume should be given The converse holds true in instances of marked hypernatremia of the body fluids

Once repair of the deficit of electrolyte and water is accomplished, the requirement for electrolyte diminishes while the water requirement to meet obligatory loss remains unchanged Adult patients may be maintained, when renal function is normal and gastrointestinal tract losses are minimal, on 40 to 50 ml per kg per day of 5 to 10 per cent glucose solution Minimal losses of electrolyte through the skin, kidney and gastrointestinal tract may be replaced by approximately 5 to 10 ml per kg per day of isotonic saline solution and 1 to 2 meq per kg per day of potassium. Potassium chloride for parenteral use may be prepared in 10 ml. sterile ampoules containing a molar solution of the salt In such a solution 1 ml contains one meq The proper quantity may be withdrawn with a sterile syringe from the ampoule and added to the daily quota of glucose and salt solution The use of small volume ampoules minimizes the possibility of over administration through error. If gastrointestinal tract losses are occurring during the post-replacement period (as is usual), provision for their replacement must be made.

It is relatively easy to discuss the theoretical therapeutics of prolonged maintenance of body fluid homeostasis with parenteral solutions. In prac-

tice, however, the problem becomes increasingly complex and clinically difficult. Most patients requiring parenteral therapy beyond a period of 3 to 4 days are those presenting such severe clinical problems as intestinal obstruction with accompanying peritoneal infection, severe degrees of diarrhea, ulcerative colitis and frank renal or hepatic disease. In such critically ill patients, salt and water homeostasis becomes progressively more unstable Often the desired urine volume and specific gravity fail to be maintained despite theoretically proper management. The kidneys may excrete excessive quantities of salt or fail entirely in this task. Hypotonicity may develop and be refractory to the administration of hypertonic saline solution. Dehydration may occur despite the apparently correct administration of water and electrolyte. Attempts to repair such dehydration often are followed by the sudden appearance of pulmonary or generalized edema. Such patients generally show evidence of progressive renal failure as manifested by rising blood non-protein nitrogen concentrations.

This failure of many patients to do well despite carefully carried out fluid and electrolyte administration has focussed attention on the fourth purpose of parenteral fluid therapy—nutrition. This apparent inability to maintain salt and water homeostasis may be attributed to nutritional failure, particularly with regard to nitrogen balance. A truly positive and sustained positive balance with a quantitatively and qualitatively adequate intake of nitrogen cannot be achieved by the organism unless the full caloric requirement is met. Protein loss can, however, be minimized by sub-optimal caloric intakes Gamble has shown that approximately 100 gm. of glucose per day for the average adult exerts maximal protein sparing effect. Increasing the quantity exerts no further effect until the full caloric intake is given. Similarly, increasing the intake of protein or amino acids on subcaloric intakes exerts no positive effect upon the nitrogen loss (16). With these observations in mind the clinician faces two alternatives in the management of the patient dependent upon a parenteral intake for nutrition Sufficient glucose may be given to provide the maximal protein sparing quantity or an attempt made to provide a full intake of both calories and protein by the intravenous route. Amino acids and protein hydrolysates may supply the protein needs when combined with glucose. About 1 gm. per kg. per day is required by adults and 2.5 to 3.0 gm per kg. for infants. To meet the adult requirement for full calories about 3 liters of 10 per cent glucose or 2 liters of 15 per cent glucose will be required Positive balances of nitrogen in experimental animals and normal men may be achieved by such means Whether this end can be accomplished in patients with infection, fever, malignancy or severe trauma is controversial The majority of evidence indicates that it cannot A major difficulty encountered in attempts to provide a complete caloric and nitrogen intake by parenteral means is the

frequent occurrence of phlebitis and sclerosis in the veins used. Such events result from the hypertonic concentrations of the solutions that must be used. The availability of fat emulsions suitable for intravenous use would obviate many of these difficulties. At the present time such preparations are in the experimental stage.

The parenteral fluid therapy of patients with complete anuria following sulfonamide therapy, incompatible blood transfusion and certain poisons, such as mercury, deserves special note. Clinical experience has clearly shown that attempts to induce diuresis by rapid expansion of the extracellular fluid by means of the intravenous administration of hypertonic glucose solutions often leads to the production of severe hypotonicity with a fatal outcome more frequently than it does to diuresis. Such patients, if managed conservatively, will remain in excellent condition for periods as long as two weeks In the great majority of instances renal function will have returned within this time and satisfactory recovery usually follows. The principles of conservative management have been described by Pratt (41) and by Strauss (42) The evaporative loss of water (10 to 20 mcg. per kg. per day) should be replaced by the intravenous or administration of 5 to 10 per cent glucose solution. Careful daily weighing of the patient facilitates estimation of the daily loss of water. Weight should be maintained constant or allowed to decrease slightly until diuresis begins Oral feeding should be avoided as it tends to produce diarrhea and vomiting in such patients.

SUMMARY

The physiology of the body fluids has been discussed in terms of the disturbances seen in clinical medicine The chemical terminology used in body fluid physiology is defined The normal relationships between size and chemical composition of extra- and intracellular fluid are discussed The means by which the acid-base equilibrium is achieved in these fluids are discussed. The pathways of water and electrolyte metabolism through the skin and lungs, the gastrointestinal tract and the kidneys are described. The obligatory nature of the skin, lung, and gastrointestinal tract expenditure is stressed in contrast with the regulatory nature of the renal loss The ways in which abnormalities in these processes lead to the production of

ticipates in these disturbances are described The repair of the body fluids by parenteral therapy is described, with emphasis on the need for considering the primary aims of such therapy—restoration of circulatory competency, repair of deficit and provision for current loss

REFERENCES

1. GAMBLE, J. L.: Chemical Anatomy, Physiology and Pathology of Extracellular Fluid. Cambridge, Harvard Univ. Press, 1951.
2. PETERS, J. P. AND VAN SLYKE, D. D Quantitative Clin. Chemistry—Vol I, Interpretations. Baltimore, The Williams & Wilkins Co , 1932.
3. PETERS, J. P. Body Water, The Exchange of Fluids in Man. Springfield, Charles C Thomas, 1935.
4. SMITH, H. W.: The Kidney, Structure and Function in Health and Disease New York, Oxford Univ. Press, 1951.
5. BUTLER, A. M. AND TALBOT, N. B.. Parenteral fluid therapy. New Eng J. Med , 231: 585-590, 621-627, 1944.
6. DARROW, D C AND PRATT, E. L.: Fluid therapy, relation to tissue composition and the expenditure of water and electrolyte, J. A. M A., 143: 365-373, 432-439, 1950
7. ELKINTON, J. R.: Water metabolism. Ann. Rev. Physiol. Stanford, Calif , Annual Reviews Inc. and the American Physiological Society, 1950.
8. GAMBLE, J. L : Companionship of water and electrolytes in the organization of body fluids (Lane Medical Lectures). Stanford Univ , Calif , Stanford Univ Press, 1950-51.
9. MARRIOTT, H. L. Water and salt depletion American Lecture Series. Springfield, Charles C Thomas, 1950
10. NEWBURGH, L H.. Significance of the body fluids in clinical medicine American Lecture Series. Springfield, Charles C Thomas, 1950
11. CLARK, W M . Topics in Physical Chemistry, 2nd ed Baltimore, The Williams & Wilkins Co , 1951.
12. SINGER, R. B AND HASTINGS, A B : An improved method of the estimation of the disturbances of the acid-base equilibrium of human blood Medicine, 27: 223, 1948
13. CONN, J. W Electrolyte composition of sweat Arch. Int Med , 83: 416, 1949
14. LOCKE, W , TALBOT, N B., JONES, H S AND WORCESTER, J. Studies on the combined use of measurements of sweat electrolyte composition and rate of sweating as an index of adrenal cortical activity J Clin Invest , 30: 325, 1951.
15. NEWBURGH, L H AND JOHNSON, N. W · The insensible loss of water Physiol Rev , 22: 1, 1942
16. GAMBLE, J L Physiological information from studies on the life raft ration Harvey Lectures 1946-47 Lancaster, Pa , Science Press Printing Co., 1947
17. COOKE, R E , PRATT, E L AND DARROW, D C The metabolic response of infants to heat stress. Yale J Biol. and Med , 22: 227, 1950
18. ADOLPH, E F. AND OTHERS *Physiology of Man in the Desert.* New York, Interscience Publishers, Inc , 1947
19. DANCIS, J , BIRMINGHAM, J R AND LESLIE, S H Congenital diabetes insipidus resistant to treatment with pitressin Am J Dis Child , 75: 316, 1948
20. WILLIAMS, R H AND HENRY, C Nephrogenic diabetes insipidus transmitted by females and appearing during infancy in males Ann Int Med , 27: 84-95, 1947.
21. WESSON, L G , JR , ANSLOW, W P , JR AND SMITH, H The excretions of strong electrolytes Bull New York Acad Med , 24: 586, 1948
22. BERLINER, R W , KENNEDY, T. J , JR AND ORLOFF, J Relationship between acidification of the urine and potassium metabolism Am J Med , 11: 274, 1951

23. MENAKER, W.: Buffer equilibria and reabsorption in the production of urinary acidity Am J Physiol., 154: 174, 1948
24. PITTS, R. F. AND LOTSPEICH, W. G : Bicarbonate and the renal regulation of acid-base balance. Am. J Physiol, 147: 138, 1946.
25 HASTINGS, A. B. AND SENDROY, J., JR · The colorimetric determination of blood pH at body temperature without buffer standards J. Biol. Chem., 61: 695, 1924
26. SCRIBNER, B. H., POWER, M. H. AND RYNEARSEN, E H.. Bedside management of problems of fluid balance. J. A. M A., 144: 1167, 1950
27. PETERS, J. P.: Problem of cardiac edema. Am J Med, 12: 66, 1951
28. FRIEDBERG, C. K · Electrolyte and fluid disturbances in congestive heart failure. New Eng J. Med, 245: 812, 1951.
29 BROWN, H., TANNER, G L. AND HECHT, H H : The effects of potassium salts in subjects with heart disease J Lab. and Clin Med, 37: 506, 1951.
30 SCHWARTZ, W. B. AND WALLACE, W. M · Electrolyte equilibrium during mercurial diuresis. J. Clin Invest, 30: 1089, 1951
31 GAMBLE, J. L ET AL Effects of large loads of electrolytes. Ped., 7: 305, 1951
32 DARROW, D C., SCHWARTZ, R, IANUCCI, J. F AND COVILLE, F.: The relation of serum bicarbonate to muscle composition J. Clin. Invest, 27: 198, 1948
33 BURNETT, C. H., BURROWS, B C AND COMMONS, R. S Studies of alkalosis I. Renal function during the following alkalosis resulting from pyloric obstruction J Clin Invest, 29: 169, 1950
34. ELIEL, L. P, PEARSON, O H AND POWSON, R. W.: Postoperative potassium deficit and metabolic alkalosis New Eng J Med, 243: 471-478, 518-524, 1950
35 ZINTEL, H A, RHOADS, J E AND RAVDIN, I S · The use of intravenous ammonium chloride in the treatment of alkalosis Surgery, 14: 728, 1943
36 FORBES, G B. AND ERGANIAN, J A Parenteral administration of ammonium chloride for alkalosis of congenital hypertrophic pyloric stenosis Am J Dis Child, 72: 649, 1946
37. CULLEN, G E The factors governing fluid therapy in the treatment of enteritis Ohio State Med J, 32: 509, 1936
38 RAPOPORT, S AND GUEST, G M The effect of salicylates on the electrolyte structure of the blood plasma I Respiratory alkalosis in monkeys and dogs after sodium and methyl salicylate, the influence of hypnotic drugs and of sodium bicarbonate on salicylate poisoning J Clin Invest, 24: 759, 1945
39 BURROWS, B A Unpublished observations
40 GOVAN, C D AND DARROW, D C The use of potassium chloride in the treatment of the dehydration of diarrhoea in infants J Ped, 28: 541, 1946
41. PRATT, E L Treatment of anuria management of patients with intrarenal lesions Am J Dis Child, 76: 14, 1948
42 STRAUSS, M B Acute renal insufficiency due to lower-nephron nephrosis New Eng J Med, 239: 693, 1948

Angiocardiography

Practical Applications

CHARLES T. DOTTER, M.D. AND ISRAEL STEINBERG, M.D.

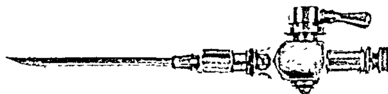
Conventional roentgenographic and fluoroscopic examination of the chest is limited by the fact that the cardiac chambers and the great vessels constitute indistinguishable components of a homogeneous shadow, the mediastinal silhouette. The situation is analogous to gastro-intestinal radiography where it is necessary to fill the lumen of the gastrointestinal tract with radiopaque barium in order to achieve the maximal diagnostic potential inherent in the roentgen ray. A practical method for the visualization of the chambers of the heart, the pulmonary circulation and the great blood vessels of the thorax was first described by Robb and Steinberg in 1938 (1). In essentials, their technic (2) is the same as that employed today. During the fourteen years since its introduction, angiocardiography has been widely used. It is the purpose of the following discussion to describe briefly the technic, emphasizing its practical diagnostic applications (3).

GENERAL DESCRIPTION OF TECHNIC

Angiocardiography consists of the roentgenographic study of the thoracic cardiovascular structures immediately following the rapid intravenous injection of a large quantity of radiopaque solution. Radiographs exposed serially at intervals following the injection reveal that the contrast substance flows (as does blood) toward the heart through the innominate vein and the superior vena cava, into the right atrium. At approximately two to three seconds after the beginning of the injection the right ventricle is filled and then the opaque solution passes out into the pulmonary arteries and their smaller branches. At about five seconds after the injection, the pulmonary veins can be visualized and then the contrast agent reaches the left atrium. Finally the left ventricle and the aorta are then filled, the cycle of opacification having lasted approximately ten seconds. Films made serially during the progress of the contrast agent outline the shape of the various cardiac chambers and vessels as they are opacified.

The angiocardiographic injection to be successful must be both large and rapidly accomplished. It is current practice to inject 50 cc. of a concentrated contrast solution into the arm vein of an adult patient within 1 to 1½ seconds. Rapidity of injection is achieved by use of a special large bore (twelve gauge) needle (fig. 1) and a special 50 cc. syringe. The needle is inserted percutaneously or, if necessary, following a cut-down. The radiographic

projection is selected depending upon the structures to be studied and the patient is positioned before the radiographic apparatus. At the time of the actual injection the patient is instructed to inspire deeply while the radiopaque solution is forcibly introduced into the vein. The x-ray exposures can be made with ordinary apparatus such as is used to make stereoscopic x-rays of the chest. With such equipment only two films can be made so that it becomes of paramount importance to predict the time of arrival of contrast agent at the desired structures. It is simpler to use more elaborate facilities equipped for automatic rapid serial roentgenography. One device, the Fairchild x-ray roll-film magazine (fig. 2) is capable of providing two exposures per second on a roll of x-ray film 9½ inches wide (4, 5, 6). With

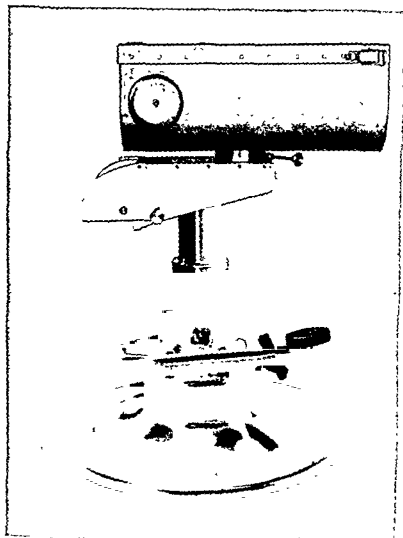


such equipment the technical features of angiocardiography are greatly simplified and the diagnostic yield is increased.

The contrast substances commonly employed for angiocardiography include diodrast 70 per cent, neo-iopax 75 per cent and urokon sodium 70 per cent. The radiopacity of each of these agents is similar. For infants and children it is customary to use approximately 1.5 cc. of the agent per kilo- [illegible] ected Injections may [illegible] ie injection is usually [illegible] ie body coupled with [illegible] with vomiting. Syn- [illegible] ema The reaction is usually over in five to fifteen minutes regardless of the contrast agent employed. In a majority of cases the vein injected becomes thrombosed The thrombus is usually firm and tender although not especially painful. Such thrombosis does not give rise to embolism and until better contrast agents are developed must be regarded as a "necessary evil"

Angiocardiography *is not* without danger At least twenty-three deaths

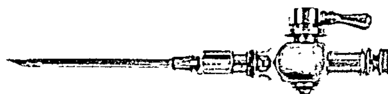
are known to have followed angiocardiographic examinations According to a survey by Dotter and Jackson (7), angiocardiographic mortality is ap-



Tl
on a
can n

proximately 0 4 per cent Most of the deaths have occurred in young children with severe congenital cardiovascular defects. At Bellevue and New York Hospital more than 2000 angiocardiographic examinations have been

projection is selected depending upon the structures to be studied and the patient is positioned before the radiographic apparatus. At the time of the actual injection the patient is instructed to inspire deeply while the radiopaque solution is forcibly introduced into the vein The x-ray exposures can be made with ordinary apparatus such as is used to make stereoscopic x-rays of the chest. With such equipment only two films can be made so that it becomes of paramount importance to predict the time of arrival of contrast agent at the desired structures. It is simpler to use more elaborate facilities equipped for automatic rapid serial roentgenography. One device, the Fairchild x-ray roll-film magazine (fig. 2) is capable of providing two exposures per second on a roll of x-ray film 9½ inches wide (4, 5, 6). With

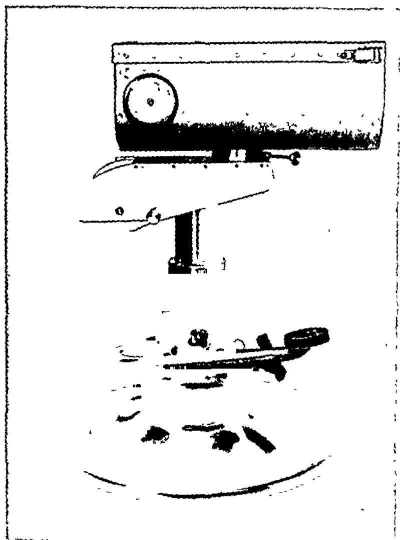


such equipment the technical features of angiocardiography are greatly simplified and the diagnostic yield is increased.

The contrast substances commonly employed for angiocardiography include diodrast 70 per cent, neo-iopax 75 per cent and urokon sodium 70 per cent. The radiopacity of each of these agents is similar For infants and children it is customary to use approximately 1 5 cc. of the agent per kilogram of body weight; for an adult 40 to 50 cc are injected Injections may be repeated if necessary The immediate reaction to the injection is usually an intense feeling of heat spreading down through the body coupled with weakness, occasionally with retching, and infrequently with vomiting Syncope may occur as may urticaria or angioneurotic edema The reaction is usually over in five to fifteen minutes regardless of the contrast agent employed. In a majority of cases the vein injected becomes thrombosed. The thrombus is usually firm and tender although not especially painful. Such thrombosis does not give rise to embolism and until better contrast agents are developed must be regarded as a "necessary evil".

Angiocardiography is not without danger. At least twenty-three deaths

are known to have followed angiocardiographic examinations According to a survey by Dotter and Jackson (7), angiocardiographic mortality is ap-



This co ... mounted on a Phil ... he device can make

proximately 0.4 per cent. Most of the deaths have occurred in young children with severe congenital cardiovascular defects. At Bellevue and New York Hospital more than 2000 angiocardiographic examinations have been

conducted without fatality. Nevertheless, angiocardiography should be done only when the results of the examination are expected to influence the treatment of the patient.

THE NORMAL ANGIOCARDIOGRAM

A major contribution of angiocardiography has been toward the understanding of the composition of the normal cardiac silhouette. Thus, the method is of value in teaching cardiac roentgenography and fluoroscopy. It has confirmed many observations based upon post-mortem study and has corrected certain misconceptions. Familiarity with normal findings is essential to the interpretation of the abnormal angiocardiogram.

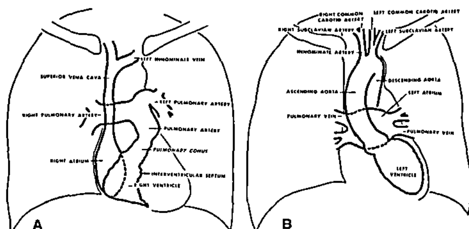


Fig 3 Idealized Angiocardiographic Outlines, Posteroanterior Projection A, right heart, B, left heart (From Dotter, C T and Steinberg, I Angiocardiography. Paul B Hoeber, Inc, New York, 1951)

The configuration of the normal heart as viewed by angiography in frontal projection is illustrated in figure 3A and B, idealized diagrammatic representations based upon the study of many normal hearts. The right mediastinal border is formed superiorly by the right innominate vein, in its mid-portion by the superior vena cava and inferiorly by the gently convex border of the thin-walled right atrium The right ventricle under normal circumstances does not extend to the right border of the heart The diaphragmatic surface of the heart is largely formed by the right ventricle The left superior mediastinal border is formed by the left innominate vein while immediately below it the aortic knob is seen to project slightly into the left lung field. Below the aortic knob, a gently convex curve is seen which is created by the main-stem pulmonary artery, often in part by its left branch and occasionally in part by the left auricular appendage. The pulmonary conus never reaches the left heart border in the frontal projection.

The lowermost convexity of the left heart border is formed by the wall of the left ventricle. In rare circumstances and particularly in older patients the ascending aorta may project to the right so as to slightly overlap the superior vena cava.

In lateral projection the upper, anterior portion of the heart silhouette is formed by the ascending aorta. Below this are the pulmonary conus and pulmonary artery while the lower anterior portion of the cardiac silhouette is formed by the wall of the right ventricle. Posteriorly, the left atrium lies above and the left ventricle below.

In the left anterior oblique projection the anterior heart border is similar to that seen in the lateral projection with the exception that the right atrium and its auricular appendage extend in variable fashion anteriorly. Occasionally the inferior vena cava may be seen close to the point where the heart overlaps the diaphragm. As in the lateral projection, the left atrium lies above with the left ventricle immediately below it

The right anterior oblique projection is rarely employed during routine angiocardiography. The posterior heart margin in this projection is formed by the atria, the left above and the right below Anteriorly and superiorly, the aortic arch and pulmonary artery (and to a lesser extent the pulmonary conus) can be seen while below and in front, the heart border is formed by the antero-lateral wall of the left ventricle.

It is well-known that the appearance of the chambers of the heart and, to a lesser extent the caliber and position of the great vessels, are altered with varying phases of the cardiac cycle (8) Usually these changes are not taken into account during angiocardiography but it is possible by means of elaborate apparatus to relate the instant of exposure to a desired phase of the cardiac cycle The information so obtained is of value in establishing standards for the normal size and wall-thickness of cardiac chambers but is not pertinent to the present discussion Measurements of the caliber of great vessels are occasionally of diagnostic value (9), particularly in the study of syphilitic aortitis.

CONGENITAL HEART DISEASE

It is in the field of congenital cardiovascular disease that angiocardiog-

nication Areas of stenosis are frequently recognizable Certain congenital cardiovascular malformations are readily recognizable clinically and do not warrant angiocardiographic study. Cardiac catheterization is more useful than angiocardiography in the diagnosis of congenital cardiovascular lesions producing left-to-right intra- or extra-cardiac shunts (e.g., septal

defects and patent ductus arteriosus). While angiocardiographic findings are often equivocal and indecisive, catheterization provides definite diagnostic information. Angiocardiography, on the other hand, is of great value in the pre-operative investigation of congenital heart disease of the cyanotic type.

Non-Cyanotic Congenital Heart Disease

Coarctation of the aorta can often be corrected by operation. Before surgery is performed the investigation of the patient suspected of having coarctation should invariably include angiocardiographic study in the left anterior oblique projection. This projection shows the aorta from the side at the angle from which it will be viewed at surgery. In the usual case there is demonstrated an abrupt narrowing of the thoracic aorta at a point variably removed but distal to the site of origin of the left subclavian artery (fig. 4). The exact anatomy of the lesion is of concern to the surgeon in allowing him to plan the proper operative attack and decide as to the advisability of vascular grafting The presence of dextro-position of the aortic arch may be easily identified by angiocardiography. In certain cases, the method has proved to be of value in the pre-operative study of constricting vascular rings about the trachea and esophagus. An anomalous course of the thoracic aorta occasionally simulates mediastinal or left hilar tumor masses, but their differentiation is made with ease by angiocardiography.

Congenital malformations causing shunting of blood from the systemic to the pulmonary circulation (left-right shunts) include the commonly encountered septal defects and patent ductus arteriosus. These lesions are frequently studied angiocardiographically In the classic interatrial defect, there is shown enlargement of the right atrium and ventricle with marked dilatation of the pulmonary arteries Frequently, but not always, the right atrium, ventricle and pulmonary arteries can still be distinguished throughout the entire period of left heart opacification This is the result of recirculation of contrast agent from the left atrium across the defect in the interatrial septum. Since a slow injection may produce a similar effect, this sign is not very reliable Patent ductus arteriosus may be suggested by recirculation of the contrast agent through the pulmonary arteries but not through the heart. In addition there may be seen a dilatation of the thoracic aorta at the site of origin of the ductus (as seen in left anterior oblique projection) (11) A recently described angiocardiographic sign of patent ductus consists of the demonstration of a defect in the contrast agent occupying the superior portion of the pulmonary artery as it is seen in frontal projection during the first few seconds following the injection (12) This is caused by a jet of non-opacified blood from the aorta entering the pulmonary artery Rarely, the communication in patent ductus arteriosus may be demonstrated by

angiocardiography, but it is more readily recognized if the contrast medium is introduced directly into the thoracic aorta (13). Cardiac catheterization is the most precise method of diagnosis of septal defects and patent ductus arteriosus; the stethoscope usually suffices for the latter.

Isolated pulmonary stenosis is commonly associated with gross dilatation of the central pulmonary arteries distal to the point of narrowing. This is a fact which has become more widely recognized since the advent of angiocardiography. Contrast visualization frequently does not reveal the exact site of the stenosis, but in demonstrating the post-stenotic dilatation it provides indirect evidence of narrowing (fig. 5). Frequently in frontal or lateral projections the actual stenosis itself can be seen. Angiocardiography occasionally can demonstrate, often suggest, but never exclude the presence of pulmonary stenosis. Again cardiac catheterization provides a more precise method of diagnosis in this form of congenital heart disease.

A relatively uncommon congenital cardiovascular lesion (which at catheterization simulates interatrial septal defect) is that of anomalous pulmonary veins draining blood from all or part of one lung into the right atrium. This lesion is readily identified by angiocardiography (14, 15).

Cyanotic Congenital Heart Disease

Clinically recognizable cyanosis due to shunting of venous blood into left heart chambers or the aorta can usually be recognized without difficulty by angiocardiography. The critical finding involves the appearance of contrast substance in the "wrong place" at the "wrong time". Immediate opacification of the aorta associated with the tetralogy of Fallot is an example. Unlike the clinical demonstration of cyanosis however, angiocardiography can be relied upon to demonstrate the site of shunting and the nature of anomaly responsible for arterial oxygen unsaturation. Congenital cardiac lesions causing cyanosis are of such a nature as to enhance angiocardiographic visualization of the malformation since right-left shunting of blood results in unusually good visualization of the chamber or vessel into which the contrast solution passes Contrast visualization has proved to be invaluable in the investigation of those congenital cardiovascular malformations which are amenable to cardiac surgery.

Fallot's tetrad consists principally of the combination of an overriding aorta and pulmonary stenosis (either in the infundibulum or at the valve area) The other two elements of the tetralogy are of necessity present if the first two occur, since an overriding aorta implies a defect between the ventricles and since pulmonary stenosis results in right ventricular hypertrophy Frontal and lateral projections suffice for adequate angiocardiographic study There is seen early and immediate opacification of the aorta (which arises from the right ventricle) and of the stenotic pulmonary artery



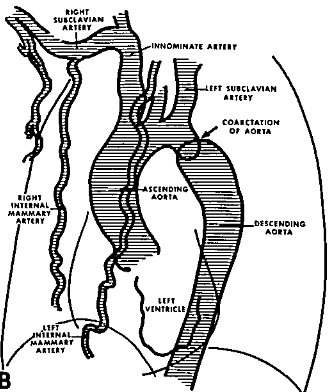


FIG 4A AND B COARCTATION OF THE AORTA.

32-year-old female Left anterior oblique angiocardiogram at 7 0 seconds Dilatation of the ascending aorta is slight. The coarctation is clearly shown 1 5 cm distal to the left subclavian artery. Collateral channels are well shown. This is the usual, so-called "adult" or operable type of coarctation (From Dotter, C T. and Steinberg, I · Angiocardiography. Paul B. Hoeber, Inc., New York, 1951)

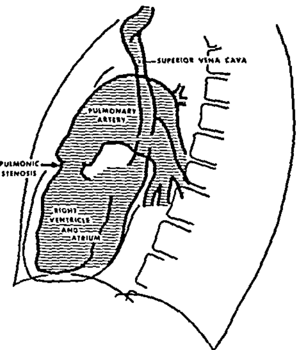
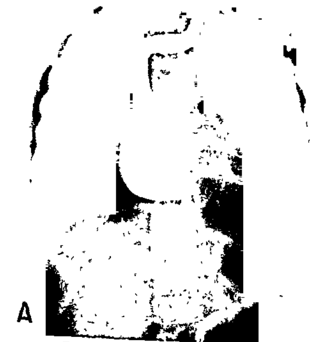


FIG 5A AND B VALVULAR PULMONARY STENOSIS

23-year-old female. Loud, harsh systolic murmur in pulmonic area Lateral angiocardiogram at 2 5 seconds There appears to be an area of narrowing at or near the pulmonary valves Poststenotic dilatation is present (From. Dotter, C T. and Steinberg, I · Angiocardiography Paul B Hoeber, Inc , New York, 1951)



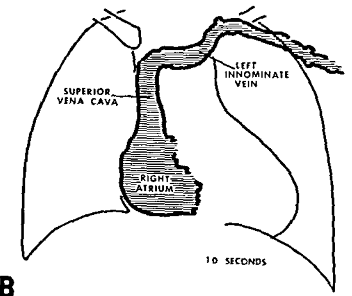


FIG 6A AND B TETRALOGY OF FALLOT DEXTROPOSED AORTA AND MODERATE PULMONARY STENOSIS.
5-year-old boy, cyanotic since 3 years Clubbing, marked restriction of activity Frontal angiocardiogram at 1 0 second, normal right atrium opacified



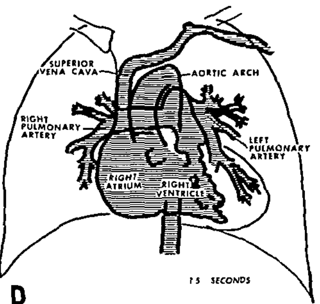
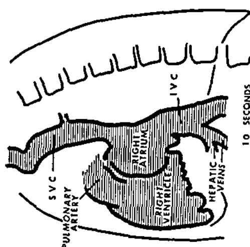


FIG 6C AND D SAME CASE AS SHOWN IN FIG. 6A AND B.
At 1 5 *seconds, there is good* visualization of a dextroposed aorta and the pulmonary arteries. An area of infundibular stenosis is visualized Pulmonary arteries are denser than aorta



E



F

Fig 6E and F. Same Case as Shown in Fig 6A to D.
Lateral angiocardiogram at 1 second Hepatic veins refluxly filled.



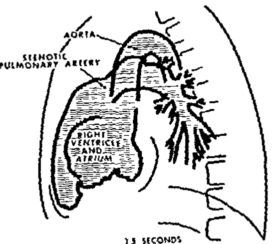


FIG. 6G AND H. SAME CASE AS SHOWN IN FIG 6A TO F.

At 1 5 seconds the aorta and narrowed pulmonary artery are seen to fill from the right ventricle. Catheterization not done. Left Blalock-Taussig operation followed by marked improvement confirming presence of pulmonary stenosis (Fig 6A to H from Dotter, C T. and Steinberg, I . Angiocardiography. Paul B. Hoeber, Inc., New York, 1951)



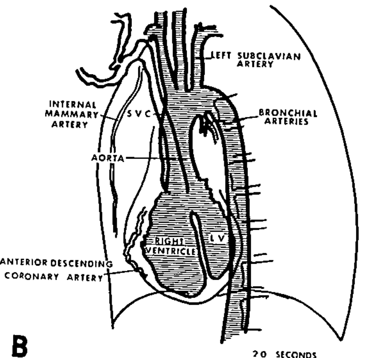


FIG. 7A AND B TETRALOGY OF FALLOT MARKED DEXTROPOSITION OF THE AORTA, EXTREME PULMONARY STENOSIS OR ATRESIA 9 year old female Cyanotic since birth, markedly diminished cardiac reserve, Mongolian idiot. Left anterior oblique angiocardiogram at 2 0 seconds There is filling of a large right and small left ventricle, the lower interventricular septum being well outlined. The aorta is large and from the lower side of the arch, two (presumably) bronchial arteries arise and pass downward toward the lung roots No pulmonary artery identifiable Obvious marked reduction of pulmonary blood flow demonstrated. (From Dotter, C. T and Steinberg, I Angiocardiography Paul B Hoeber, Inc , New York 1951)

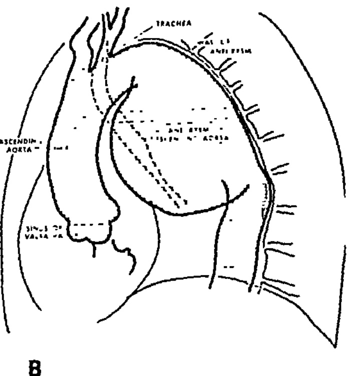


Fig 8A and B. Aneurysm of Descending Aorta
63-year-old female A Left anterior oblique angiocardiogram showing opacification of aorta and large saccular aneurysm of descending aorta. Sinuses of Valsalva well shown. B Tracing of A, aorta and aneurysm shaded. (From: Dotter, C. T. and Steinberg, I · Angiocardiography. Paul B Hoeber, Inc, New York, 1951)

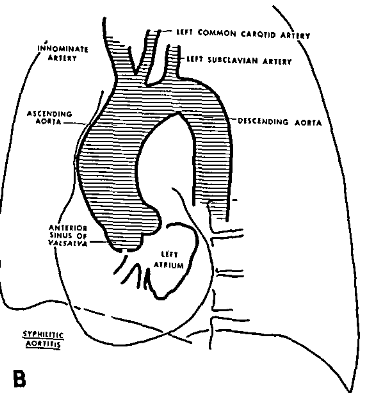


Fig 9A *and* B Uncomplicated Syphilitic Aortitis.
56-year-old male Untreated syphilis of 30 years' duration No evidence of aortic insufficiency Blood pressure, 135/75. A Left anterior oblique angiocardiogram at 8 5 seconds The midascending aorta is dilated to 42 mm. B · Tracing of A, aorta and brachiocephalic arteries shaded (From Dotter, C T and Steinberg, I · Angiocardiography Paul B Hoeber, Inc , New York, 1951)



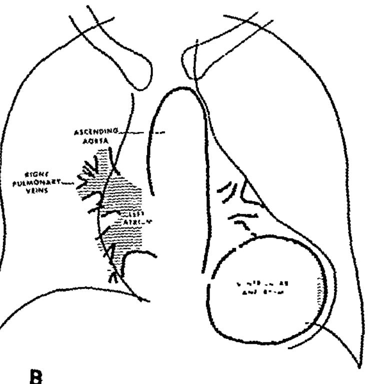


FIG 10A AND B. SACCULAR VENTRICULAR ANEURYSM.

29-year-old Negro male who had severe anterior *chest pain* and prostration 3 years before angiocardiography A Frontal angiocardiogram at 10 5 seconds. A large thin-walled saccular aneurysm arising from the left ventricle is opacified B: *Tracing of A, left heart, aorta,* pulmonary veins, and aneurysm are shaded. (From: Dotter, C. T. and Steinberg, I.: Angiocardiography. Paul B Hoeber, Inc , New York, 1951)

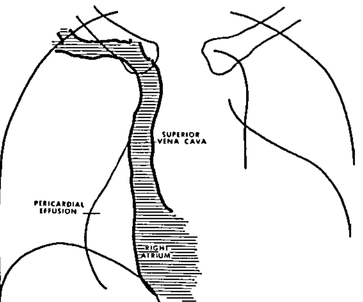


FIG 11A AND B PERICARDITIS WITH EFFUSION.

56-year-old female A Frontal angiocardiogram at 2 0 seconds There is a 3 5 cm space between the opacified right atrium and the margin of the cardiac silhouette B Tracing of A, superior vena cava and right atrium shaded Subsequent to angiocardiography, over 400 cc of straw colored fluid was removed from the pericardium The cause of the effusion was not established.

(fig. 6). The pulmonary stenosis is occasionally not directly identifiable but its presence may be inferred from the demonstration of distorted, small, or poorly filled pulmonary arteries. The actual stenosis is demonstrated in



FIG 12 PULMONARY EMPHYSEMA.
52-year-old female with pulmonary emphysema and pulmonary hypertension (33/21 mm Hg at cardiac catheterization) Frontal angiocardiogram at 3 0 seconds shows central pulmonary arterial enlargement due to the hypertension The peripheral pulmonary arterial branches are widely separated and poorly seen due to the emphysema The right ventricle is not demonstrably enlarged, the interventricular septum is concave (From Dotter, C T and Steinberg, I Angiocardiography Paul B Hoeber, Inc , New York, 1951)

about half of the cases studied Aside from establishing or confirming the clinical diagnosis of tetralogy of Fallot, angiocardiography aids by demonstrating the position of the aortic arch and its brachiocephalic arteries, and the size and position of the pulmonary arteries, information of practical value to the surgeon. Accordingly contrast visualization should be performed routinely prior to the Blalock-Taussig operation.

Transposition of the great blood vessels does not always produce a char-



Fig 13A and B Pulmonary Arteriovenous Fistula.

31-year-old female with cyanosis since childhood A soft systolic murmur was heard over the left lower lobe posteriorly Hemoglobin was 17 5 Gm A Frontal angiocardiogram at 2 5 seconds There is diffuse varicoid dilatation of the terminal branches of the pulmonary arteries to the posterolateral portions of the left lower lobe These vessels can be seen to communicate directly with pulmonary veins B Lateral angiocardiogram at 2 5 seconds A film at 5 0 seconds showed aortic opacification, evidence of the arteriovenous shunting of unoxygenated blood The patient's husband refused to allow her to be operated upon although told of the benefit to be anticipated from lobectomy (From Dotter, C. T. and Steinberg, I : Angiocardiography. Paul B Hoeber, Inc , New York, 1951)

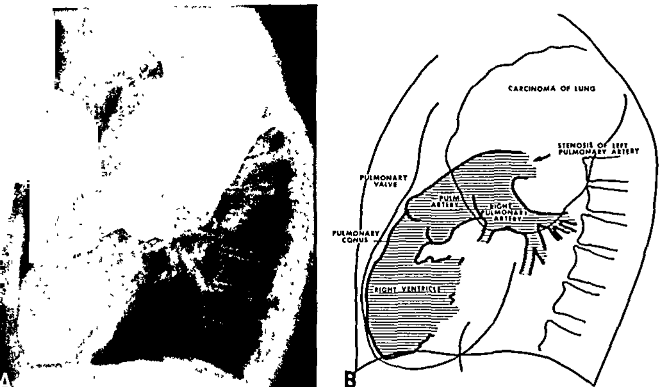


FIG 14A AND B CANCER OF THE LUNG—OCCLUSION LEFT PULMONARY ARTERY.
61-year-old male with 6 months' history of cough showed almost complete occlusion of the left main bronchus on bronchoscopy and a large rounded mass in the left upper lobe. Lateral angiocardiogram at 3 0 seconds. Complete occlusion of left pulmonary artery at bifurcation. Angiocardiographic diagnosis of inoperability confirmed by exploratory thoracotomy and autopsy. (From: Dotter, C T and Steinberg, I Angiocardiography Paul B Hoeber, Inc , New York, 1951.)

acteristic cardiac silhouette on conventional roentgenographic or fluoroscopic examination. Due to rotation of the large blood vessels, the superior mediastinal vascular pedicle is often narrow in frontal projection and unusually broad in lateral or left oblique views. The pulmonary vascular shadows may be normal or increased, but are diminished if pulmonary stenosis is present. Angiocardiography reveals that the aorta arises from the right ventricle but fails to show early filling of the pulmonary artery. It is sometimes difficult to distinguish between transposition of the great blood vessels and the so-called "pseudo-truncus arteriosus" (16). In the latter malformation, the pulmonary artery is atretic or too small to be visualized (fig. 7). The diagnosis of transposition is facilitated by lateral angiocardiograms where the aorta is seen to describe a wide-open curve through the thorax. If an associated patent ductus arteriosus is present it may be directly revealed Occasionally the pulmonary arteries are seen to opacify from the left ventricle at a period following that of aortic opacification.

Pulmonary stenosis associated with an interatrial septal defect (often a patent foramen ovale) may simulate the tetralogy of Fallot on angiocardiographic study By virtue of the pulmonary stenosis however right ventricular end-diastolic pressure is usually elevated with resultant elevation of the right atrial pressure. As a consequence, shunting of blood from the right to the left atrium occurs, a phenomenon readily identifiable by angiocardiography There is seen to be enlargement of the right atrium, direct spread of contrast agent from the right to the left atrium with early opacification of both the aorta and the pulmonary artery The latter is usually stenotic and may show post-stenotic dilatation In further contrast to Fallot's tetrad, the pulmonary artery in this condition is emptied of contrast substance more slowly, while the aorta clears immediately It is sometimes possible to demonstrate the exact site of the pulmonary stenosis, a fact of great surgical significance since in this condition a direct attack on the area of stenosis (Brock operation) can be planned

Tricuspid stenosis or atresia is readily diagnosed clinically through the combination of cyanosis and left axis deviation The angiocardiogram demonstrates right-left interatrial shunting, a large left ventricle and a rudimentary or absent right ventricle (17) Pulmonary stenosis may or may not be demonstrated. In common with pulmonary stenosis associated with patency of the foramen ovale, tricuspid stenosis or atresia usually is accompanied by right atrial enlargement and opacification of the hepatic veins by reflux filling

ACQUIRED CARDIOPULMONARY DISEASE

Most forms of acquired heart disease do not often require angiocardiography, other diagnostic methods sufficing For example, arteriosclerotic

coronary artery disease is profitably studied by the electrocardiogram while hypertension requires but the use of a sphygmomanometer. Rheumatic heart disease lends itself to highly accurate clinical and roentgenologic methods of diagnosis while angiocardiography deserves no role in its routine study. On the other hand, in certain specific types of acquired cardiac or pulmonary disease angiocardiography has proved to be an indispensable diagnostic aid.

Syphilitic Aneurysm and Aortitis

One of the most quickly appreciated and more dramatic applications of contrast cardiovascular visualization was in the differential diagnosis between intrathoracic aneurysm and tumor (18, 19). It has long been known that aneurysms may fail to show pulsation on fluoroscopic examination while tumors in contiguity with great blood vessels may seemingly exhibit expansile pulsation Few thoracic surgeons of wide experience have not at least once performed thoracotomy only to discover an aneurysm instead of the suspected tumor With the aid of angiocardiography, this situation need never occur On angiocardiographic examination, the aneurysm fills with contrast agent while the parent vessel is demonstrably dilated and distorted (fig. 8). If tumor is present the aorta is unremarkable and the mass does not opacify. Save in rare cases with intraluminal thrombus formation the angiocardiographic identification of aneurysm is readily made even by the uninitiated. Contrast visualization has proved itself to be of value in the demonstration of dilatation, tortuosity, and elongation of the thoracic aorta associated with syphilitic aortitis (20). It is capable of demonstrating very early dilatation in the aorta, even in the absence of clinical signs (21) Dilatation of the mid-ascending aorta to above forty millimeters in caliber is abnormal and usually accompanies syphilitic aortitis (fig 9) Moderate aortic dilatation may be the result of hypertension, aortic valvular insufficiency of any cause, or coarctation of the aorta and in the presence of these lesions the diagnosis of syphilitic aortitis cannot be based solely on the demonstration of dilatation

Rheumatic Heart Disease

Angiocardiography has added to present-day roentgenologic knowledge regarding the abnormal cardiac silhouette associated with rheumatic mitral stenosis (22) It has become apparent that the prominence in the mid-portion of the left border of the so-called "mitralized" heart reflects the presence of 1) enlargement of the left atrium and its auricular appendage and 2) dilatation of the pulmonary artery and its left branch The pulmonary conus does not participate in forming this portion of the cardiac silhouette In lateral projection it is readily seen that the principal alterations

in the cardiac silhouette are the result of enlargement of the left atrium. Enlargement of this chamber actually displaces the right ventricle and atrium anteriorly, causing these structures to encroach upon the retrosternal space in such a way as to simulate right ventricular enlargement (3). Aside from the elucidation of the foregoing facts, angiocardiography has proved to be of little practical clinical use in the study of rheumatic heart disease. It is currently being employed to study the size of the left atrium prior to and following operations on the mitral valve.

Arteriosclerotic Heart Disease—Ventricular Aneurysm

The coronary arteries are small and inconstantly visualized with the result that their study by angiocardiography is not practical. Elongation, tortuosity, and increased wall-thickness of the thoracic aorta are strikingly *demonstrated by opacification but can be easily recognized on conventional* roentgenographic study. One of the significant complications of coronary artery disease, ventricular aneurysm, may be diagnosed readily and demonstrated strikingly by angiocardiography (23). Contrast films reveal opacification of an out-pocketing or saccular dilatation of the left ventricle, a pathognomonic finding (fig 10). The distinction between aneurysm and massive scar formation following myocardial infarction is chiefly a matter of extent, degree of bulging and localization of the weakened area. Following myocardial infarction the angiocardiogram frequently reveals marked thinning of the antero-lateral wall of the left ventricle, a finding of diagnostic significance. Contrast visualization has successfully been employed to distinguish between ventricular aneurysm and para-cardiac tumor. Buckling of the innominate artery may simulate right superior mediastinal tumor, an illusion readily dispelled by angiocardiography (24).

Dissecting Aneurysm

Suspicion of dissecting aneurysm of the aorta is usually based upon the coexistence of hypertension and severe penetrating chest pain unassociated with evidence of coronary arterial occlusion Unfortunately the diagnosis frequently is not established prior to post-mortem examination. With angiocardiography the abnormal anatomy can be revealed easily (25). Films made in frontal or left anterior oblique projection demonstrate abnormal narrowing of the aortic lumen at the site of dissection coupled with a marked thickening of the aortic wall *It is sometimes possible to demonstrate partial* or complete occlusion of the brachiocephalic vessels consequent to the dissection. Dissecting aneurysm has been identified angiocardiographically in patients who denied previous chest pain

Pericardial Effusion

The presence of fluid within the pericardial sac may be mimicked closely by an enlarged, dilated and poorly pulsating heart. Angiocardiography makes the differentiation simply (26). Films in frontal projection timed to reveal the right atrium demonstrate an increased area of non-opacification between the cavity of the right atrium and the right lung (fig. 11). Left heart films reveal a similar increased area between contrast substance within the left ventricle and the adjacent lung. The method allows the identification of small amounts of fluid and also demonstrates the otherwise hidden cardiac contour. The differentiation between pericardial effusion and mediastinal or paracardiac tumors can readily be made.

Pulmonary Disease and Pulmonary Heart Disease

Modern concepts of the pathogenesis of pulmonary heart disease stress a relationship between anoxia due to pulmonary disease and pulmonary hypertension (27). Cor pulmonale is a term usually reserved for such cases of pulmonary hypertension when failure of the right ventricle has developed. Although angiocardiography is not the most satisfactory method for demonstrating right ventricular hypertrophy (unipolar electrocardiography is superior), it affords a demonstration of the presence and degree of pulmonary arterial dilatation, an early change in pulmonary heart disease. Cardiac catheterization and pulmonary function studies are the most useful methods for the study of abnormal cardiopulmonary physiology.

Vascular changes in the lung field are seen commonly as a result of chronic pulmonary diseases such as tuberculosis (28), pulmonary fibrosis and long standing bronchiectasis An estimate of the function of a given lung or portion of it can be obtained by the study of pulmonary vascular opacification on angiocardiography. In emphysema, the pulmonary vessels are seen to be widely separated and poorly filled peripherally while they may be dilated centrally (fig 12) Vascular changes produced by pulmonary fibrosis are non-specific. In "fibrothorax", thickened pleura, atelectatic lung and the heart occupy one side of the chest. The contour and position of vascular structures cannot be made out in the conventional roentgenogram, but they can be seen with angiocardiography. Angiocardiography has demonstrated clearly a reduction in pulmonary arterial flow on the side of pneumothorax (29) and thoracoplasty (30). By outlining areas of rehabilitatable lung in bullous emphysema, contrast visualization has been of practical value in the selection of patients for surgical resection

Pulmonary arteriovenous fistula is one operable pulmonary lesion capable of causing cyanosis, polycythemia, and disability. Its presence may be suspected on the basis of clinical findings, its location may be identified with

reasonable certainty on the conventional roentgenogram. Angiocardiography affords far more complete anatomical information and should invariably precede surgical resection (fig. 13).

INTRATHORACIC NEOPLASM

It is reasonable to predict that in the years to come, extensive diagnostic applications of angiocardiography will be made in two fields, congenital heart disease and tumors of the mediastinum and lungs. Through its ability *to delineate the vascular structures*, angiocardiography significantly expands the diagnostic and prognostic investigation of intrathoracic neoplasms. It provides anatomical information of value to the surgeon contemplating operative removal of such lesions

Superior vena caval block, readily identifiable by conventional clinical and laboratory methods, may be delineated anatomically by means of opacification of the superior vena cava and its tributaries (31). The extent and exact site of the block may be demonstrated as well as the resultant collateral venous channels. The appearance of the block may be of diagnostic significance in that external pressure defects and the stenosing effects of surrounding tumor are occasionally distinguishable It is, however, often impossible to differentiate between block caused by a malignant tumor and that resulting from mediastinitis with superior vena caval thrombosis.

Mediastinal Tumors

One of the principal hazards in the operative removal of a mediastinal mass lies in the fact that the surgeon is able to see only the front, and to a lesser extent the sides of the mass he is dissecting If the surgeon has available information from angiocardiography regarding the nature of the vascular structures immediately behind the tumor, he is better able to avoid perforation of a blood vessel and its often catastrophic consequences Contrast visualization, by delineating sites of extension of the tumor mass, enables the surgeon to complete his exploratory procedure more rapidly (32). In general, benign mediastinal tumors produce external pressure defects or simple dislocation of blood vessels whereas malignant neoplasms are apt to cause occlusion with marked distortion It is occasionally possible to identify the presence of malignancy on the basis of vascular changes visualized by angiocardiography. Pre-operative angiocardiographic study obviates the necessity for exploration in the differentiation of mediastinal tumor and aneurysm In our opinion, angiocardiography should be performed routinely prior to surgery in any suspected mediastinal tumor

Carcinoma of the Lung

Since by contrast visualization one can distinguish between the vascular changes produced by a malignant and a benign process, the procedure is of

value in the investigation of carcinoma of the lung (33, 34). Although not infallible, in many instances of pulmonary carcinoma, contrast visualization can establish the diagnosis and give information regarding operability. Peripheral tumors usually produce no recognizably significant changes in the pulmonary arteries although minor segmental alterations do occur. Central tumors often cause partial or even complete occlusion of the major pulmonary arteries, particularly the left (fig. 14). Evidences of inoperability in carcinoma of the lung include: 1) partial or complete occlusion of the left pulmonary artery within one to two centimeters of its point of origin, 2) partial or complete occlusion of the right pulmonary artery proximal to its site of bifurcation, 3) metastatic occlusion of the mediastinal veins, 4) pericardial invasion. Many malignant tumors show no vascular evidence of their invasiveness and mediastinal extension may occur without causing demonstrable angiocardiographic abnormality.

By indicating the anatomy of the tumor, angiocardiography may shorten the time of exploration in lung cancer By proceeding directly to the site of suspected mediastinal metastasis the surgeon may establish the nature and extent of the disease without tedious preliminary dissection Angiocardiography may save the patient an unnecessary surgical procedure by demonstrating extension of the tumor beyond the limit of surgical resectability. As a rule, however, angiocardiographic findings alone cannot be considered sufficient cause for withholding surgery Non-neoplastic conditions may result in vascular changes similar to those produced by cancer of the lung.

It is predicted that in certain types of lung cancer, particularly those with central, hilar or mediastinal masses, angiocardiography will become a routine part of the pre-operative investigation

REFERENCES

1. Robb, G P and Steinberg, I Practical method of visualizing chambers of the heart, pulmonary vessels and great blood vessels in man J Clin Invest., **17**: 507, 1938.
2. Robb, G. P and Steinberg, I Visualization of the chambers of the heart, the pulmonary circulation, and the great blood vessels in man A practical method. Am J Roentgenol, **41**: 1–17, 1939
3. Dotter, C T and Steinberg, I Angiocardiography Paul B Hoeber, Inc, New York, 1951
4. Dotter, C T, Steinberg, I and Temple, H L Automatic roentgen-ray roll-film magazine for angiocardiography and cerebral arteriography. Am J Roentgenol, **62**. 355–358, 1949
5. Smith, D C and Dotter, C T Automatic x-ray roll-film magazine for angiocardiography technical considerations X-Ray Tech, **22**. 365–369, 1951.
6. Dotter, C T and Steinberg, I Rapid serial contrast angiography Angiology, **2**: 173–183, 1951
7. Dotter, C. T and Jackson, F S Death following angiocardiography Radiol., **54**: 527–534, 1950

8. FREDZELL, G , LIND, J., OHLSON, E. AND WEGELIUS, C.: Direct serial roentgenography in two planes simultaneously at 0 08 second intervals. Am. J. Roentgenol , 63: 548–558, 1950
9. DOTTER, C. T. AND STEINBERG, I.: The angiocardiographic measurement of the normal great blood vessels. Radiol., 52: 353–358, 1949.
10 DOTTER, C. T AND STEINBERG, I . Angiocardiography in congenital heart disease Am. J. Med., 12: 219–237, 1952
11. STEINBERG, M. F., GRISHMAN, A. AND SUSSMAN, M. L : Angiocardiography in congenital heart disease. III Patent ductus arteriosus. Am J Roentgenol., 50: 306–315, 1943.
12 GOETZ, R. H.: A new angiocardiographic sign of patent ductus arteriosus. Brit Heart J , 13: 242–246, 1951.
13 CASTELLANOS, A. AND PEREIRAS, R . Retrograde or countercurrent aortography Am. J. Roentgenol , 63: 559–565, 1950
14. DOTTER, C T , HARDISTY, N M AND STEINBERG, I. Anomalous right pulmonary vein entering the inferior vena cava· report of two cases. Am J Med Sci , 218: 31–36, 1949.
15 GRISHMAN, A , POPPEL, M H , SIMPSON, R S AND SUSSMAN, M. L.· The roentgenographic and angiocardiographic aspects of (1) aberrant insertion of pulmonary veins associated with interatrial septal defect and (2) congenital arteriovenous aneurysm of the lung Am J Roentgenol , 62: 500–508, 1949
16 COOLEY, R N , BAHNSON, H T AND HANLON, C. R Angiocardiography in congenital heart disease of cyanotic type with pulmonic stenosis or atresia I *Observations on the tetralogy of Fallot and "Pseudo-Truncus Arteriosus"* Radiol , 52: 329–346, 1949
17. COOLEY, R N , SLOAN, R D., HANLON, C. R AND BAHNSON, H T Angiocardiography in congenital heart disease of cyanotic type II Observations on tricuspid stenosis or atresia with hypoplasia of the right ventricle Radiol , 54: 848–868, 1950
18 STEINBERG, I AND DOTTER, C T. The differentiation of mediastinal tumour and aneurysm value of angiocardiography Brit J Radiol , 22: 567–572, 1949
19 SUSSMAN, M. L. The differentiation of mediastinal tumor and aneurysm by angiocardiography Am J Roentgenol , 58. 584–589, 1947
20 STEINBERG, I , DOTTER, C. T , PEABODY, G , READER, G , HEIMOFF, L AND WEBSTER, B The angiocardiographic diagnosis of syphilitic aortitis Am J Roentgenol , 62. 655–660, 1949
21 PEABODY, G E , READER, G G , DOTTER, C T , STEINBERG, I AND WEBSTER, B Angiocardiography in the diagnosis of cardiovascular syphilis Am J Med Sci , 219: 242–248, 1950
22. GRISHMAN, A , SUSSMAN, M L AND STEINBERG, M F Angiocardiographic analysis of the cardiac configuration in rheumatic mitral disease Am J Roentgenol., 51: 33–43, 1944.
23. DOLLY, C H , DOTTER, C T AND STEINBERG, I Ventricular aneurysm in a 29 year old male studied angiocardiographically Am Heart J., 42: 894–899, 1951
24 HONIG, E I , STEINBERG, I AND DOTTER, C T Innominate artery angiocardiographic study Radiol , 58: 80–87, 1952
25. GOLDEN, A. AND WEENS, H S The diagnosis of dissecting aneurysm of the aorta by angiocardiography report of a case Am Heart J , 37: 114–118, 1949
26. WILLIAMS, R G. AND STEINBERG, I The value of angiocardiography in establishing the diagnosis of pericarditis with effusion Am J Roentgenol , 61: 41–44, 1949

27. Riley, R L, Himmelstein, A, Motley, H. L, Weiner, H M and Cournand, A: Studies of the pulmonary circulation at rest and during exercise in normal individuals and in patients with chronic pulmonary disease. Am. J. Physiol., **152**: 372-382, 1948.

28. Steinberg, I., McCoy, H. I. and Dotter, C. T.: Angiocardiographic findings in pulmonary tuberculosis. Dis. Chest, **19**: 510-520, 1951.

29. Steinberg, I., McCoy, H. I. and Dotter, C T: Angiocardiographic findings in artificial pneumothorax. Am Rev. Tuberc, **62**: 353-369, 1950.

30. McCoy, H. I., Steinberg, I and Dotter, C. T. Angiocardiographic findings in thoracoplasty, artificial pneumoperitoneum and phreniclasia. J. Thorac Surg, **21**: 149-158, 1951.

31. Roberts, D J, Jr, Dotter, C. T and Steinberg, I: Superior vena cava and innominate veins. angiocardiographic study. Am J. Roentgenol, **66**: 341-352, 1951.

32 Steinberg, I, Dotter, C T, and Andrus, W. deW: Angiocardiography in thoracic surgery Surg, Gyn & Ob, **90**: 45-59, 1950

33. Dotter, C T., Steinberg, I. and Holman, C. W: Lung cancer operability. Angiocardiographic study of 53 consecutive proved cases of lung cancer. Am. J. Roentgenol., **64**: 222-238, 1950

34. Steinberg, I. and Dotter, C T. Lung cancer Angiocardiographic findings in 100 consecutive proved cases. A.M A Archives of Surgery, **64**: 10-19, 1952.

Portal Hypertension

MARY ANN PAYNE, M.D., Ph.D. AND
CHARLES G. CHILD, III, M.D

Massive hematemesis and melena associated with portal hypertension constitute a serious medical and surgical problem. The initial episode of hemorrhage may be fatal or the patient may survive only to have a recurrence. The source of hemorrhage has repeatedly been shown to be the esophagogastric varices which develop in direct response to increases in portal pressure. In the past, numerous surgical operations have been devised to lower this abnormally high pressure Because of their magnitude and seriousness, these procedures were never widely applied in the treatment of patients with this syndrome. In recent years, however, adequate preoperative and postoperative techniques have been developed for the physiological support of these critically ill patients. Through these ancillary measures as well as by improvements in vascular surgery, portal venous decompression has become an important feature in the management of patients with esophagogastric hemorrhage who today can be assured with reasonable certainty that they need not succumb to an episode of hemorrhage.

While portal hypertension frequently develops as a sequel to cirrhosis, increases in portal pressure are by no means limited to hepatic disease. Excessively high pressures in the portal venous system may be associated with any pathological process that produces a partial or complete block to portal blood flow Where this occurs, regardless of cause or associated conditions, massive esophagogastric bleeding may occur. Such hemorrhage may be a more serious threat to life than the primary disease Any patient who has had a hemorrhage from esophagogastric varices requires, therefore, detailed attention to this aspect of his illness It is the purpose of this chapter to review the background of the syndrome, to consider some of the difficulties in diagnosis, and to evaluate the current methods of treatment

HISTORICAL

Although in Europe, Stahl as early as 1698, Koeniff in 1751 and Raikem in 1848 were apparently aware of most of the clinical manifestations of portal hypertension, Power in this country was one of the first to describe accurately massive bleeding from esophagogastric varices In 1840, he performed a careful autopsy on a colored man who had had repeated attacks of melena and who died in shock following one such severe episode Power described a plexus of veins in the lower esophagus and noted a large ulcera-

tion of one of these thin-walled vessels from which he could easily express residual blood. He wrote, "Varicose veins of the esophagus I have never met with before, nor have I found any similar case recorded; they may sometimes exist to a lesser degree, but I doubt whether their presence to such an extent, and death caused by their rupture, be not something as yet unheard of in pathological anatomy." This autopsy report then continued with a detailed description of the stomach, heart, and other organs, but no mention was made of the liver or of collateral circulation connecting the portal with the systemic venous beds. Power did not speculate as to the cause of the varices or of their rupture He attempted, however, to establish his case as the first on record showing the relationship between esophageal varices and bleeding. Fauvel described a similar case in 1858 In 1860-61 a comprehensive two volume treatise on liver disease was published by Frerichs who referred to Fauvel's report as the only case of bleeding from esophageal veins that he had ever known He concluded that hemorrhage must represent a highly unusual occurrence even though varicosities might be present. Despite an exhaustive discussion of liver disease, Frerichs failed to associate varices and hemorrhage with the liver or the portal vein.

In Paris about 1900, Gilbert and Weil, Villaret and Pichancourt, by their studies upon abdominal ascites, firmly established portal hypertension as a concomitant of cirrhosis Their interests lay primarily in the formation of ascites and urinary suppression rather than in esophagogastric hemorrhage. Preble, however, also in 1900 presented a clear and concise report of 60 cases of fatal gastrointestinal hemorrhage in patients with cirrhosis of the liver This was a comprehensive review and included only cases with autopsy reports Esophageal varices were present in 80 per cent of the cases and in more than half of these the site of erosion was demonstrated. Preble believed that hemorrhage in cirrhosis occurred either from varices developing as a result of mechanical obstruction to the portal circulation or as an expression of acquired hemorrhagic diathesis He felt, however that the majority of these gastrointestinal hemorrhages were from varices.

Preble was well aware of the relationship between intrahepatic obstruction to portal blood flow and the development of esophageal varices He reviewed the possible anastomotic channels between the portal and systemic circulation which he believed opened up in response to increasing increments of pressure in the portal system He thought that the anastomoses between the coronary veins of the portal system and the systemic veins of the esophagus were particularly liable to varix formation because of their proximity to the liver The interference with portal blood flow was attributed by Preble to the loss of elasticity of the liver as it became cirrhotic.

An experimental approach to the cause of increased portal pressure in portal cirrhosis was made by Herrick in 1907 He perfused human normal

and cirrhotic livers obtained at autopsy to study the relationship of portal vein pressure to hepatic artery pressure. In perfusing normal livers, when the portal pressure was maintained at 10 millimeters of water, an increase of 40 millimeters in arterial pressure was necessary in order to effect a rise of 1 millimeter in portal vein pressure However, in a cirrhotic liver, a comparable rise of 1 millimeter of portal pressure resulted from every 6 millimeters rise of arterial pressure. Herrick concluded, therefore, that in the cirrhotic liver there existed a freer communication between the arterial and portal circulations.

Herrick also believed there was an increased arterial supply to the cirrhotic liver. In perfusing normal livers with portal pressure maintained between 10 and 20 milliliters of water, portal flow always appreciably exceeded arterial flow. This was not a new observation, for it was well known that normally the portal vein supplies more blood to the liver than does the hepatic artery. In perfusing cirrhotic livers, however, it was necessary to raise portal pressure above 40 millimeters of water in order to render portal flow greater than arterial. Thus, Herrick reasoned that portal flow *was less than arterial flow in the cirrhotic liver except in far advanced* cirrhosis where the elevated portal pressure kept normal or nearly normal amounts of blood flowing through the liver. He attributed the increase in portal pressure to two factors: 1) more direct communication between the arterial trees and the portal veins, and 2) increased volume of blood supplied to the liver by the hepatic artery. Herrick believed that both factors operated to transmit arterial pressure more freely to the venous bed He found no evidence of actual obstruction to portal flow from the fibrosis of the cirrhotic liver.

McIndoe, using the same perfusion technique, was unable to duplicate these observations of Herrick McIndoe thought that fibrosis gradually interfered with the blood supply to the parenchyma of the liver and that a large part of the fluid perfused through the system escaped through the collaterals. Dock in 1942 repeated these experiments and combined the observations of both Herrick and McIndoe He postulated a reciprocal relationship between hepatic artery and portal venous flow proportional to the degree of both arterial pressure and intrahepatic resistance. Dock pointed out that results might be expected to vary with the type of cirrhosis and the stage of the disease. His non-alcoholic cirrhotic liver preparations did not show any increase in hepatic arterial perfusibility while significant increase in flow was demonstrated in the alcoholic cirrhotic liver He suggested that failure to differentiate various types of cirrhosis might account for the variation in Herrick's and McIndoe's results.

In spite of the many studies devoted to this problem, the precise explanation for portal hypertension in cirrhosis has proved elusive indeed.

Even today the pathological processes involved in cirrhosis are in themselves poorly understood. Where portal hypertension develops in a person with a normal liver, this phenomena can be studied without the physiological abnormalities of cirrhosis.

Guido Banti, an Italian physician and teacher of anatomy, first focused medical attention upon a group of patients who had little or no evidence of primary liver disease, but who presented signs known today to be those of increased portal pressure. In his original work on splenic anemia published in 1882, he cited the history of a 16-year-old girl who developed a large spleen and progressive anemia. At the onset of her illness she appeared to have a normal liver. Although she was "stricken with hepatic cirrhosis" two years later, Banti pointed out that this case was probably not ordinary Laennec's cirrhosis He suggested that she had a primary splenomegaly and later developed hepatic cirrhosis In the course of the next few years, he saw ten more cases that he thought fitted this picture. In 1894, Banti reported the original case and three additional ones under the heading of "Splenomegaly with Hepatic Cirrhosis". These four patients were selected by him because they were followed over a long period of time and ultimately to the autopsy table. It is important to consider in some detail the disease which Banti described, for much confusion has arisen over its exact nature.

Banti described his patients as "young adults" although actually his four cases ranged in age from 15 to 54 years. He laid great stress on the history, pointing out that he was unable to elicit any of the factors so frequently associated with Laennec's cirrhosis, such as dietary deficiency, alcoholism, infections, poisons, or toxins He observed these patients over a span of years and described a progression of symptoms which he also incorporated into his diagnostic criteria Banti believed that anemia and splenomegaly antedated the appearance of ascites. During this early period, which lasted one to four years or more, these patients were relatively asymptomatic. Weakness or left upper quadrant discomfort was often the only complaint. The progression of the anemia and the appearance of digestive disturbances characterized what he considered the intermediate stage. Mild liver involvement could be detected at this time and oliguria was a prominent feature The entire intermediate stage lasted only a few months, progressing rapidly to the ascitic or terminal phase. Here all the signs of liver failure were apparent. The patient usually failed rapidly, developed jaundice, and became comatose. Death occurred in seven to eight months. It is interesting that even in his later papers Banti does not mention hematemesis in relation to this clinical picture but stresses "liver failure" as the cause of death Banti repeatedly stated that the cause of this disease was unknown, he thought that the primary site of involvement was the spleen Furthermore, he believed that the periarterial

fibrosis, the so-called fibro-adenie in the Malpighian bodies was distinctive and pathognomonic.

In 1900, Osler related his experiences with fifteen patients whom he classified as having splenic anemia. He credited Griesinger with coining the term "splenic anemia" and pointed out that he had no personal knowledge of the condition described by Banti. Osler concluded from his study that until more was known about this syndrome it was useful to group together patients presenting splenomegaly and anemia without lymphatic enlargement He recognized that hematemesis was one of the features of the disease but did not offer any specific explanation for its occurrence.

From the time that Banti first presented his syndrome, scattered reports appeared of similar cases in which portal or splenic vein thrombosis had been demonstrated at autopsy. Rommelaere in 1903 called attention to the relationship between primary splenomegaly and septic thrombosis of the splenic and portal veins Edens, in 1908, described portal thrombosis associated with a clinical picture similar to Banti's syndrome Cauchois in the same year took issue with Banti's theory of a primary splenic origin for the syndrome and reported a number of cases which he believed demonstrated portal splenic thrombosis as the initiating event. Warthin, in 1910, and Eppinger, a few years later, emphasized the clinical similarity between Banti's disease and the splenomegaly that develops secondary to chronic splenic vein thrombosis During the next fifteen to twenty years, numerous observations were reported in the literature stressing this relationship

Gradually, various types of extrahepatic obstruction became associated with the syndrome described by Banti. In 1921, Hart stated that sclerosis of the portal vein could produce such a clinical picture. Klemperer, in 1928, wrote a comprehensive review of cavernomatous transformation of the portal vein and showed by careful documentation its relationship to the picture of Banti's disease Larrabee, in 1934, attributed the syndrome to obstruction of the splenic vein

In 1934, John McMichael reviewed congestive splenomegaly from the pathological viewpoint under the heading of hepatolienal fibrosis He described periarterial fibrosis and dilated venous sinuses in the spleen wherever an acute inflammatory condition of the liver was present. He thought these splenic abnormalities could be intensified but could not be caused by increased portal pressure McMichael considered these vascular changes to be the same as the fibro-adenie of Banti and had his microscopic slides reviewed by Aschoff who had seen Banti's preparations Aschoff agreed with McMichael that the two lesions were identical

The present concept of portal hypertension and its broader relationships to cirrhosis as well as to the syndrome described by Banti grew out of the work of the Spleen Clinic at Presbyterian Hospital, New York City.

Rousselot, in 1936, and Thompson, Caughey, Whipple, and Rousselot, in 1937, measured the pressure in the splenic vein and found a marked increase in splenic vein pressure in cases presenting Banti's syndrome. They suggested that portal hypertension was an important factor in the production of this syndrome. In 1940, Rousselot reported 15 cases of Banti's syndrome and listed a variety of extrahepatic lesions responsible for the obstruction. He stated that his group believed that Banti's syndrome was due to obstruction of portal or splenic blood flow.

Whipple, in 1945, reviewed portal hypertension in relation to the earlier work in cirrhosis and to the more recent work of his own group on Banti's syndrome. He suggested that patients with portal hypertension be divided into two mains groups:

Group 1. Patients with *intrahepatic portal block* due to cirrhosis.

Group 2. Patients with *extrahepatic portal block* due to occlusion of the portal vein or one of its main tributaries.

This classification and terminology is generally accepted today. It has been exceptionally useful in establishing a clear understanding of portal hypertension in all its various clinical manifestations.

Almost from the time physicians, surgeons and pathologists became concerned with the clinical and pathological picture of portal hypertension, laboratory investigators interested in the physiology of the liver began to study the complex relationships of the hepatic circulation. To consider even briefly all of the experimental work that has been performed upon the blood flow in this organ is far beyond the scope of this chapter. Only those physiological studies, therefore, which are relevant to a reasonably complete understanding of portal hypertension in man will be reviewed.

Delimited as it is by two capillary beds, the portal venous system is unique. It is not surprising, therefore, that the portal vein early became the object of laboratory investigation. As early as 1856, 1863, and 1877, Oré, Schiff, and Claude Bernard interested themselves in the effect of sudden interruption of portal blood flow. The rabbit, cat and dog did not survive portal vein ligation by more than an hour or so. Many explanations were advanced to account for this phenomenon, namely, generalized toxemia, anemia, or liver failure. In 1877, Lautenbach, working in Schiff's laboratory in Geneva described a syndrome appearing in mammals after portal ligation characterized by a great tendency to sleep, a decreased arterial pressure and death. It was ascribed to a poison which Lautenbach believed was not destroyed when portal blood was prevented from passing directly through the liver. Eck apparently did not accept this or other theories and was stimulated to devise the fistula which now bears his name. In his original article, Eck wrote, "It was established that the blood of the portal vein can, without any danger to the body, be diverted directly into the

general circulation." In 1875, Solowieff showed that the dog could survive *completely portal vein occlusion provided its several branches were ligated* individually and in stages.

Although these early investigators must have noticed the intense venous congestion appearing in the dog's mesentery after portal occlusion, they paid little attention to the accompanying changes in splanchnic venous pressures. *The primary interest during this early period was whether or not the* animal survived portal occlusion. Not until 1934 was the cause of death following sudden portal occlusion in the common laboratory animals discovered At this time, Elman and Cole and Boyce demonstrated that the cat and the dog died in shock after sudden occlusion of the portal vein. *This they proved was due to decrease of effective circulating blood volume.* Quite literally, these animals bled to death into their portal venous systems.

It is difficult to determine the exact beginnings of interest in experimentally produced portal hypertension. In 1894 Bayliss and Starling, in association with their classical experiments upon capillary pressure and blood flow, *occluded the portal vein of the dog* many times and noted that upon first trying this experiment portal pressure rose to heights too great to measure with their manometers They did state, however, that this rise must have been well over 60 to 80 centimeters of a 25 per cent solution of magnesium sulfate Bayliss and Starling also pointed out the marked *congestion of the liver and the increased lymph flow that resulted from* occlusion of the outflow tract of the liver—the hepatic veins or the inferior vena cava above the liver A few years later Chiari described hepatic venous obstruction in man. This is usually fatal, and of rare occurrence In 1909, Schmid reported numerous experiments largely concerned with *the effect of various drugs, such as adrenalin, upon portal pressure.* Many of these produced sharp elevations in the portal pressure of the dog

During the early 1900's, great interest was expressed in the relationship between the hepatic arterial and portal venous flow In 1914 McLeod and Pearce demonstrated that occlusion of the hepatic artery reduced hepatic flow at least 30 per cent while occlusion of the portal vein reduced this flow some 60 per cent.

Herrick, McIndoe, and Dock all realized the importance of increased portal blood pressure in cirrhosis They employed perfusion experiments upon human cirrhotic livers because a satisfactory method had not been devised for reproducing cirrhosis in experimental animals. Although today numerous chemicals, infectious agents, and deficient diets are known to produce cirrhosis in the rat, rabbit and dog, these forms of liver disease cannot convincingly be compared with human cirrhosis. Furthermore, great difficulty has been experienced in producing portal hypertension by any of these methods It was not until 1939 that Rousselot was able to produce in

dogs a cirrhosis which was associated with significantly elevated portal pressure and splenomegaly. This he accomplished by multiple injections of silica into the portal vein over a period of 7 to 37 months.

The first experimental work upon portal hypertension without cirrhosis is that of McMichael. While working in Wilkie's laboratory in Edinburgh in 1932, McMichael became interested in the fact that, "The condition of portal hypertension . . . can occur in the absence of such gross and late changes as are found in a hob-nailed liver." This investigator postulated that, "If portal hypertension is present in the early stages of liver disease, it must depend more on an alteration of physiological conditions of the portal circulation than on gross anatomical changes." McMichael then repeated many of the previously reported experiments upon the effect of adrenalin, vasopressin, and other drugs upon portal pressure. He did not, however, at this time reach any definite conclusion in explanation of how portal hypertension was produced without liver disease Two years later, however, he demonstrated that in man portal hypertension can exist without cirrhosis.

Rousselot (1940) must be given credit for establishing extrahepatic block as a cause for portal hypertension in man. This fact, as well as the demonstration by Rousselot, Whipple, Blakemore, Linton and others that esophagogastric hemorrhage due to portal hypertension can be controlled by portal decompression, has stimulated a number of investigators to attempt to produce this syndrome in laboratory animals. These efforts have failed almost uniformly. Gradual occlusion of the portal vein in the dog causes so many collaterals to appear about the site of occlusion that any transient increase in portal pressure subsides within a few weeks. Bollman in several stages occluded both the dog's portal vein and vena cava above the liver and attempted to force the development of collateral circulation through the esophageal veins by sclerosing other collateral channels The esophageal veins were enlarged but definite varices were never produced Accessory collaterals always developed to replace those occluded. Child and his associates have shown that the *Macaca Mulatta* monkey, unlike many other laboratory animals, may survive sudden and complete portal occlusion uneventfully. Furthermore, although a marked elevation in portal pressure to 35 to 60 centimeters of saline occurs immediately after occlusion, this subsides to almost normal levels within a week or so. Apparently, as in other animals, the collateral circulation which develops in the monkey is adequate to decompress the portal system effectively. In man, Child has also demonstrated that the portal vein can be resected in the course of pancreaticoduodenectomy for carcinoma of the pancreas. Two such patients followed for six and ten months respectively failed to develop esophageal varices Although the evidence is far from

complete, it appears at this time that significant degrees of sustained portal hypertension cannot be produced in man or in animals by mere occlusion of the portal vein.

The relationships of the hepatic artery to portal pressure and blood flow have interested laboratory investigators for almost as long a period as has the portal vein itself. In 1909, Heberer and Baudouin both demonstrated that common laboratory animals died following interruption of the hepatic artery. Whereas sudden portal occlusion was fatal within an hour or two, division of the hepatic artery produced death from hepatic necrosis in a matter of days No real advance was made in determining the cause of the liver necrosis until Markowitz, Rappaport and Scott demonstrated nearly fifty years later that when dogs with a properly ligated hepatic artery are given penicillin for 7 days postoperatively, they usually survived indefinitely and the liver was not abnormal. Dogs protected by penicillin survive because this drug prevents the proliferation of anerobes which are normally present in the dog's liver. Child and his associates, on the other hand, have demonstrated that in the *Macaca Mulatta* monkey such bacteria are normally absent. This animal survives hepatic artery ligation without antibiotics.

The reason for the current intense interest in the hepatic artery is found in Reimhoff's claim that in man ascites and esophagogastric hemorrhage can be treated effectively by ligation of this vessel distal to the origin of the gastroduodenal artery The importance of this claim is great indeed. for ligation of the hepatic artery is a simple operation compared with portal decompression by a veno-venous shunt Today in clinics throughout the country, the effectiveness of lowering portal pressure by a combination of hepatic and splenic arterial ligation is being studied

Madden* has demonstrated the following meager falls in pressure in the portal vein in patients with cirrhosis after ligating the hepatic and splenic arteries (see table 1).

At the New York Hospital the hepatic artery has been temporarily occluded in five patients with cirrhosis There has been marked variation in the effect on portal pressure In one patient hepatic arterial occlusion produced a drop in portal pressure of 10 cm of saline, in another patient a fall of but 2 cm was observed, in three patients no effect at all was produced.

Insofar as man is concerned, it seems apparent at this time that the hepatic artery cannot be ligated safely if the liver is normal (Grant, Fitts, and Ravdin). In the presence of cirrhosis, however, current evidence indicates that the hepatic artery can be interrupted successfully. Not only

* Personal communication, 1951

does it appear certain that patients with this disease survive occlusion of the hepatic artery, but they may even be relieved of portal hypertension to a therapeutic degree. Just as other laboratory studies on portal hypertension have been handicapped by the lack of a satisfactory form of cirrhosis in animals, so probably will clarification of the relationship of the hepatic artery to increased portal pressure be delayed.

In addition to the animal studies on portal vein and hepatic artery, other aspects of hepatic blood flow have been carefully investigated. Their exact relationship to sustained portal hypertension is not entirely clear. For instance, Kirschner, Hooker and Shearer produced ascites and portal hypertension in the dog by gradual occlusion of the vena cava above the liver.

TABLE 1

Direct recording in mm of saline

	CASE 1	CASE 2	CASE 3	CASE 4
Initial pressure	399	482	425	410
Ligation of hepatic artery proximal to gastroduodenal artery	375 (24)*	460 (22)*	412 (13)*	398 (12)*
Combined ligation of hepatic and splenic arteries	344 (31)†	421 (39)†	405 (7)†	— ‡
Total decrease in portal venous pressure	55	61	20	

* The fall in pressure after ligation of hepatic artery

† The additional fall after ligation of splenic artery.

‡ Splenic artery not ligated.

Simonds and Brandes, and Child, working on the dog and monkey respectively, produced a great increase in portal pressure by occlusion of the hepatic veins All these animals, however, died within minutes to hours after sudden hepatic vein occlusion. Knisley, Wakim, and Mann have studied extensively intrahepatic arteriovenous dynamics Although their observations have contributed to a better understanding of the anatomical and functional relationships between the portal vein, the hepatic artery and the hepatic veins, they have not served to clarify the cause of portal hypertension

ETIOLOGY OF PORTAL HYPERTENSION

In considering the etiology of portal hypertension, the causative factors involved can best be discussed under two headings: Portal hypertension due to intrahepatic block, and portal hypertension due to extrahepatic block

Intrahepatic Portal Block

The term "intrahepatic block" is used by many authors interchangeably with cirrhosis. This term implies chronic liver disease with concomitant slow development of scar tissue. Many agents have been implicated in the cause of cirrhosis including alcohol, malnutrition, various industrial chemicals, and severe infections. It is generally accepted today that almost any insult to the liver which continues for a long period of time eventually results in permanent pathological changes such as are found in cirrhosis. Because fibrosis is one of the prominent features of the cirrhotic liver, it has long been held that it was this process itself which actually produced the portal hypertension by blocking portal blood flow. Kelty, Baggenstoss, and Butt have recently examined this concept critically, basing their opinions upon wax reconstruction of a cirrhotic liver, and came to the conclusion that it is not the fibrosis which obstructs blood flow but rather that it is regenerating nodules of hepatic cells. These, they believe, impinge upon the lumens of the portal radicles to a sufficient degree to impair the passage of portal blood through the liver If this concept be correct, portal hypertension in cirrhosis develops as a result of hepatic regeneration rather than fibrosis. This explanation, of course, is attractive, particularly when it is realized that not all patients with cirrhosis develop clinically significant hypertension. It is known from Patek's study of Laennec's cirrhosis that portal hypertension occurs in about one-third of the cases. Aherns reported portal hypertension in over half of a group of patients with biliary cirrhosis.

Portal hypertension, then, may be associated with any chronic process in the liver which leads to fibrosis and regeneration of liver tissue. Repeated heavy infestation with Schistosoma mansoni or Schistosoma japonicum often leads to cirrhosis of the liver Portal hypertension may develop in from three to five years after infestation In addition, extensive granulomas form in the mesenteric venous bed during the period of ovopositing

the spirochete in the production of cirrhosis Although the tertiary phase of syphilis with hepar lobatum and portal hypertension is rare, the history of such a patient has been reported in whom a portacaval shunt was performed for massive hematemesis (Lazzari and Rack). Occasionally portal hypertension is encountered in association with relatively rare diseases. Patients with hemochromatosis and an associated cirrhosis may develop portal hypertension Mino has reported a patient with Boeck's sarcoid with extensive liver infiltration who developed portal hypertension and on whom a shunt was performed successfully.

Extrahepatic Obstruction

Either septic or bland thrombosis of the portal, superior mesenteric, or splenic veins is the commonest cause of extrahepatic block. This may be acute from injury, or chronic, secondary to slowly progressive portal phlebosclerosis. Trauma to the epigastrium and the left upper quadrant is a well recognized cause of portal thrombosis The first patient reported by Whipple was a young policeman with traumatic ruptures of the pancreas He developed a pancreatic cyst, and then over a three-year period the classical syndrome of congestive splenomegaly developed. Portal obstruction may also occur as the result of extension of thrombosis from one or another of the mesenteric veins. Mesenteric thrombosis following an operation within the pelvis, though rare, does occur. Thrombosis of the splenic vein following splenectomy is capable of producing this type of portal venous obstruction.

Mesenteric pylephlebitis is one of the major causes of portal thrombosis. Langdon Brown collected twelve such cases and in 1901 wrote an excellent description of this pathological process. Appendicitis, biliary tract infection, intestinal tuberculosis, puerperal sepsis, ulcerative colitis and diverticulitis have all been reported as sources of septic thrombi. Omphalitis may be an important factor in the production of an extrahepatic block. The history of this infection can often be elicited from children with portal obstruction Generalized infections have been implicated in portal bed thrombosis. Mallory reports a case following mastoiditis, Warthin one following pneumonia, and Smith and Farber one following scarlet fever.

Phlebosclerosis of the portal vein and its tributaries is most difficult to evaluate as a cause of extrahepatic obstruction. These vascular changes led Banti to postulate that a toxin was liberated from the spleen which caused progressive injury to the vessels of the portal system Other investigators have reported extensive mesenteric sclerosis both with and without thrombosis. It has never been decided whether this process is a primary disease or whether it occurs secondary to high portal pressure Patients with polycythemia may develop portal hypertension and esophagogastric hemorrhage As the red cell count rises, blood viscosity increases and the rate of blood flow diminishes This, together with the increased number of cells, predisposes these patients to thrombosis not only of small but also of large vessels, and typical extrahepatic block may be produced The same factors which produce the extrahepatic block make these patients exceptionally poor operative risks Not only is thrombophlebitis a common postoperative complication, but the success of the shunt may be jeopardized by thrombosis In polycythemia bleeding from esophagogastric varices may be massive and may make operation imperative. The following case illustrates the problems and some complications involved.

Case No 1, B W NYH #545 435 Age 67 In 1946, at age 63, this man had an enlarged spleen, enlarged liver, and an elevated red blood cell count. A diagnosis of polycythemia vera was made He was bled at intervals of six to eight weeks for almost three years In the spring of 1949, he had his first esophageal hemorrhage. Because he bled twice more in the next four months he was admitted to the hospital. Hepatosplenomegaly persisted The diagnosis of polycythemia vera was confirmed Esophageal varices were demonstrated by x-ray. The patient was considered a poor operative risk and was discharged However, he continued to have repeated esophageal hemorrhages and was re-admitted in October 1950

He was a poorly nourished man of 67 years. His blood pressure was 110/70 There was no cyanosis or icterus There was a suggestion of ascites The edge of the liver could be felt three finger-breadths below the costal margin, the spleen five finger-breadths He had bilateral inguinal herniae and an umbilical hernia There was slight pitting edema of the ankles

Laboratory data included a hemoglobin of 8 grams; red cell blood count of 4.3 million, white cell count of 9,600, serum protein of 6 3 grams per 100 ml with an albumin of 4 4 grams per 100 ml, bilirubin 0 8 milligrams per 100 ml, alkaline phosphatase 5 units, thymol turbidity 3 units, cephalin flocculation 8 units, bromsulfalein retention 7 per cent, prothrombin time 18 5 seconds

Despite the poor operative prognosis, he was explored on December 27, 1950, through a right thoraco-abdominal incision Pressure in the superior mesenteric vein was 41 centimeters of saline After many hours of tedious dissection the portal vein was found thrombosed and replaced by many smaller, dilated venous channels Many of these were also thrombosed A portacaval anastomosis was impossible, and the incision was closed At this time it was planned to attempt a splenorenal shunt at another operation However, a massive hematemesis occurred on the seventh postoperative day It was controlled by the Sengstaken Blakemore balloon He developed a pleural effusion, renal failure, and died 19 days after operation The esophageal balloon had been in place for 12 consecutive days with an intraluminal pressure of 20 to 25 millimeters of mercury

Post mortem examination disclosed hyperplastic bone marrow, thrombotic occlusion of the portal vein, old coronary artery thrombosis, and thrombosis of the pulmonary vein In addition, an acute ulcer 3 centimeters in diameter was found in the esophageal wall It had eroded through the entire thickness of the esophageal wall anteriorly and exposed the tracheal rings

So-called cavernomatous transformation of the portal vein is one of the most interesting phenomena seen in portal hypertension Pick was convinced that this lesion was a hemangioma, and described neoplastic invasion of the wall of the portal vein Others have thought it a congenital anomaly, particularly where found in children Most pathologists consider the mass of dilated veins a sequel to reorganization and recanalization of a thrombosed portal vein On occasion, remnants of the original portal vein can be found by careful search of the cavernomatous mass. The end stage of this thrombotic process may be a fibrous cord surrounded by an extensive collateral circulation.

Congenital anomalies account for a small percentage of cases of portal bed block. Mahoney reported two patients with congenital stricture of the portal vein, both of whom had experienced massive hemorrhage from

esophageal varices by the age of four. Smith and Farber described obstruction to portal blood flow by persistent fetal valves in the portal vein with thrombosis behind the valve leaflets. They suggest that this may not be an uncommon cause of thrombosis in children, for the portal system and its tributaries contain valves in the fetus. These normally persist for only a short time after birth.

Extrinsic compression of the portal or splenic veins may lead to obstruction. Benign and malignant tumors, cysts, particularly those of the pancreas, inflammatory masses or even aneurysms of the splenic artery have produced mild degrees of portal hypertension. Malignant tumors of the pancreas, stomach, gall bladder and common bile duct frequently obstruct major vessels of the portal bed. These tumors may invade the wall of the vessel and actually fill the lumen. Primary hepatomas invade the portal vein and the hepatic vein and also grow within the lumen of these vessels. Significant portal hypertension in relation to malignant growths is rare, and its presence is usually unimportant in the over-all prognosis of the patient

THE DIAGNOSIS OF PORTAL HYPERTENSION

Esophagogastric Bleeding

Esophagogastric hemorrhage may be the first and only evidence of portal hypertension Since cirrhosis of the liver is the main cause of portal hypertension, it is in this group of patients that hemorrhage most frequently occurs. Esophagogastric bleeding was the initial symptom in 10 per cent of the 444 patients with uncomplicated Laennec's cirrhosis reported from the Mayo Clinic by Douglass Hematemesis and melena occurred in 32 per cent at some time in the course of their illness These figures are comparable with those from the series reported earlier by Preble, Ratnoff and Patek and Rolleston.

The identification of the source of hemorrhage in a patient who has had a massive gastrointestinal hemorrhage may be relatively simple or most difficult Massive hemorrhage in a patient long under treatment for cirrhosis presents few problems in diagnosis The clinical history and studies point to the varices as the most probable source of bleeding Any young individual with an unheralded hematemesis should be suspected of having bled from varices occurring secondary to an extrahepatic block In contrast to such patients where the correct diagnosis is all but obvious are those in whom a given hemorrhagic episode might be due to any one of the recognized causes of gastrointestinal bleeding To establish varix as the source of bleeding is hard Patients in the fifth or six decades whose hemorrhage is from varices secondary to unrecognized cirrhosis offer many problems in differential diagnosis.

In this connection, Ricketts has recently pointed out the high incidence of "latent" portal cirrhosis unrecognized in most routine investigations. In his series of 50 such cases diagnosed by biopsy, 8 per cent had esophageal bleeding as the first manifestation of disease. Zamcheck suggested that the bromsulfalein test be used to distinguish the patient with cirrhosis who is *probably bleeding from varices from the patient with a normal liver* probably bleeding from other causes. He found only an occasional elevation of bromsulfalein retention in patients with bleeding peptic ulcer and in these the test returned to normal during observation. In contrast, he did not find a normal bromsulfalein in any patient bleeding from esophageal varices. Regardless of a previously normal liver function tests, liver function at the time of bleeding in a patient with cirrhosis almost invariably is abnormal. It should be borne in mind, however, that the establishment of the diagnosis of cirrhosis does not necessarily imply that the bleeding is *coming from varices For instance, pyloric and duodenal ulcers are more* frequently found in patients with cirrhosis than in the normal population Snell found ulcers in 10 per cent of patients with cirrhosis.

Many patients enter the hospital after the acute episode of bleeding is over or respond to blood replacement within a few hours. Careful clinical and roentgenographic examination usually establish the diagnosis. Others enter the hospital with an acute bleeding episode and continue to bleed massively while under observation. Even under these circumstances, prompt roentgeongraphic examination may disclose an ulcer, carcinoma or varix. Out of White's 400 cases of bleeding admitted to the Boston City Hospital, varices were demonstrated in 64 patients

Patients in whom the source of bleeding can not be demonstrated by the usual diagnostic procedures are the greatest challenge. Twenty-six per cent of the cases reported by Schiff in 1944 fell into this category. In 5 per cent of White's large series, the cause of bleeding could not be established.

In massive esophagogastric hemorrhage, all diagnostic measures may fail. Here, the physician is justified in employing tamponade as an aid in diagnosis. By applying pressure directly to the areas most commonly involved by varices it may be possible to infer the site of hemorrhage Esophagoscopy may be helpful in an attempt to visualize varices during bleeding. It is usually, however, difficult under emergency circumstances

Collateral Circulation

The single most important diagnostic sign of portal hypertension is collateral circulatory channels connecting the portal with the systemic bed When the pressure in the portal bed rises significantly above normal, the smaller connecting radicles between the portal and the systemic circulation

open and more and more portal blood bypasses the liver through systemic collaterals. In intrahepatic obstruction these characteristically develop in the following areas:

1) At the two ends of the gastrointestinal tract where glandular epithelium unites with squamous epithelium; namely, the gastric cardia and the anus. Esophagogastric varices form submucosally at the site of anastomosis between the coronary system and the azygos, hemi-azygos and diaphragmatic system of veins. Conventionally, these varices have been thought to be limited to the esophagus but recent experience gained in demonstrating these dilated vessels roentgenographically has emphasized that they may occur as frequently in the cardia of the stomach. The anastomosis between the superior hemorrhoidal vein of the portal circulation and the middle and inferior hemorrhoidal vein of the caval circulation form hemorrhoids which only occasionally become significant in the management of the patient with portal hypertension.

2) At the umbilicus. Here the partially obliterated embryological circulation in the falciform ligament connects the portal bed with the veins of the anterior abdominal wall. As this collateral circulation develops huge venous channels conduct blood to the systemic circulation through the branches of the lateral thoracic and epigastric veins.

3) At the sites within the abdomen where the gastrointestinal tract and its appendages developmentally become retroperitoneal (veins of Retzius).

4) Occasionally an extensive collateral circulation may develop where abnormal viscera have become adherent to the parietal peritoneum as a result of an inflammatory process.

Where the obstruction to portal blood flow lies outside of the liver numerous anastomotic channels develop in the immediate vicinity of the occluded portal or splenic veins. These partially circumvent the block and portal blood gains access to the liver almost directly The components of this type of collateral circulation are the deep cholecystic veins, the epiploic veins of the gastric omentum, the hepatocolic and hepatorenal veins, and the accessory veins of Sappey.

Recently, Edwards has reviewed the entire subject of communications between the portal and systemic circulations. In addition to confirming the observations of Eighteenth Century anatomists, this investigator discusses in detail the potential adequacy of these collaterals in portal venous obstruction. He emphasizes particularly the functional importance of the anastomoses about the umbilicus, the spleen, the left kidney, and the rectum

As interest in portal hypertension has increased, numerous efforts have been made to introduce refinements in diagnosis designed to prove the presence of portal block. Circulation time from the rectum to the lungs has

been measured by means of ether instilled into the lower colon. Where this is prolonged, it is used as evidence of an obstructed portal flow (Newman). Bean has endeavored to measure portal pressure pneumatically by observing the pressure at which the sigmoidal mucosa blanches. Because of their accessibility, the distended veins on the anterior abdominal wall have been employed by a number of investigators as aids to establish a diagnosis of portal block. Various sugars and proteins, after oral ingestion, have been shown to appear earlier and in greater concentration in the abdominal veins than in those in the antecubital area (Sherlock and Walshe; Billings and DePree; Bean, Franklin, Embick, and Daum; Blondheim). Several investigators (Davidson, Gibbons, and Faloon, Levy and Burch) have found the pressure in the abdominal veins elevated in patients suspected of having portal hypertension. Bloom has re-emphasized the venous hum heard over the epigastrium in patients with cirrhosis. These indirect methods of measurement are of little value as a routine guide in diagnosis or therapy, however, because of the great variability and individuality of pattern in the venous bed.

In addition, it must be remembered that increased portal pressure is not the only factor concerned in the alteration of the collateral circulation of the abdominal wall. Pelvic tumors, ascites and gas cause demonstrable rises in pressure in the adbominal veins. Injury or inflammation of the femoral or epigastric veins produces a superficial collateral pattern over the lower abdomen identical with that seen in portal hypertension

The most significant clinical aspect of collateral circulation between the portal and systemic circuit is the presence of esophagogastric varices Their demonstration is accepted as positive evidence of abnormally elevated pressure in the portal bed The use of thin, adhesive contrast mixtures brings out the mucosal pattern by x-ray sufficiently well to identify the small "bean-shaped" radiolucent areas characteristic of varices. Special positioning and the use of Valsalva maneuver has been found useful in this clinic in demonstrating varicosities of the esophagus The pattern of the normal gastric mucosa is so varied that varices in the cardia of the stomach are extremely difficult to detect At the present time, the radiographic demonstration of esophagogastric varices is the single most important diagnostic procedure in establishing the presence of portal hypertension.

Splenomegaly

Splenomegaly is not pathognomonic of portal hypertension. Indeed, of the 1180 cases of splenomegaly studied by Whipple, evidence of portal hypertension was present in only 174 cases or 14 per cent. This small group included all cases of portal hypertension caused by intrahepatic obstruction

such as cirrhosis or extrahepatic block. The remaining 86 per cent were in patients with anemia, hemolytic jaundice, Hodgkin's disease, chronic myeloid leukemia, polycythemia, and purpura They had no sign of portal hypertension. Many of the patients came to operation, and portal pressures were measured (Thompson). In patients with portal bed block, portal pressures ranged from 225 to over 500 millimeters of water. In the control group with large spleens but without portal obstruction, pressures were 70 to 290 millimeters of water. The large spleen did not increase pressure in the portal bed

In 1948, Moschowitz extensively reviewed the pathogenesis of splenomegaly in portal hypertension and submitted his reasons for believing that the elevated pressure in the portal system is the only factor involved He also expressed the opinion that the duration of the hypertension was a more important factor in increasing the intensity of the lesions seen in this type of splenic enlargement than the actual height of the pressure. While increased portal pressure may contribute to splenic enlargement, there is some evidence that this is not the only factor concerned Cameron has demonstrated that splenic enlargement in rats appeared in response to exposure to carbon tetrachloride even when the splenic connection to the portal bed had been severed He did this by an ingenious experiment in which he first transplanted the spleen to the abdominal wall thereby removing all direct connections with the portal circulation The animals were then exposed to carbon tetrachloride and hepatic necrosis induced. The splenic transplants consistently enlarged regardless of the position in portal or the systemic circulation. It would appear, therefore, that splenic enlargement was independent of any increased portal pressure in this case. The enlargement of the spleen may be interpreted as a response either to the carbon tetrachloride itself, acting directly on the spleen, or as a secondary response to the products liberated from the degenerating liver cells

The anemia, leukopenia, and thrombocytopenia associated with the syndrome of portal hypertension are more properly related to the large spleen rather than to the portal bed block Although the exact mechanism whereby splenomegaly produces this hematologic picture is not known, it is believed to be a manifestation of hyper-function of this organ. For this reason the currently popular use of the term "hypersplenism" seems justified

THE TREATMENT OF HEMORRHAGE FROM ESOPHAGOGASTRIC VARICES

Although massive hemorrhage from esophagogastric varices was recognized as an important and dramatic clinical entity about the middle of the last century, little progress was made in its management before 1900 Paracentesis was performed merely for relief of ascites Splenectomy,

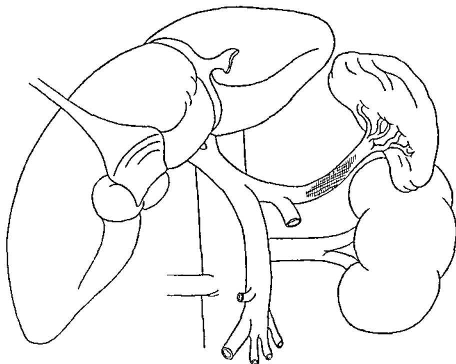
practiced for splenic anemia as early as 1865 by Spencer Wells and in 1875 by Pean was undertaken primarily to relieve pain and discomfort caused by the large spleen. When Eck in 1877 published his widely quoted suggestion that the venous fistula which now bears his name might prove useful in relieving the manifestations of portal bed block, he had the treatment of ascites in mind. The original omentopexy which Talma and Morison advocated independently, 1898–1903, was designed to increase the blood supply to the atrophic liver rather than to promote drainage of the portal venous system.

During the period from 1900 to 1915, serious efforts were made to elucidate the etiological factors involved in cirrhosis of the liver; efforts at therapy were directed primarily toward the relief of ascites For instance, Vidal in 1900 made an Eck fistula upon a patient with advanced ascites in whom an omentopexy was impossible. About 1910, de Martel and Lenoir made Eck fistulae in two patients with ascites. Both died in anuria shortly after operation. In 1912, Rosenstein made an Eck fistula in a patient with severe ascites and reported that after operation, paracentesis had to be performed less frequently. During the latter part of this period, surgeons apparently became discouraged with large vessel anastomosis and confined their efforts to lesser venous shunts between portal and systemic veins (Gunn 1911, ovarian to portal, Meursing 1912, spermatic to splenic; Bogoras 1913, superior mesenteric to vena cava). Success was so meager, however, that direct decompression of the portal bed fell into relative disfavor Neither minor nor major venous anastomoses were attempted again for many years

From 1915 to the late 1930's, no real advances in therapy were made Only indirect methods of portal bed drainage were practiced. Enthusiasm for one or another variation of the Talma-Morison omentopexy was expressed sporadically, but review of the reported results (Mayo, Grinnell, Cates) fails to provide convincing proof of any significant relief from ascites or hemorrhage Splenectomy continued to be practiced widely during this period for splenic anemia or so-called Banti's disease (Mayo, Pemberton, Andrus and Holman) With regard to the prevention of recurrent hemorrhage, two impressions prevailed. First, a few patients could be completely cured by removal of the spleen These good results must have been obtained in patients whose varices were caused by splenic vein occlusion alone (fig. 1) Second, it was also appreciated that those with cirrhosis, if they survived the operation, were seldom protected against recurrent hemorrhage. As the result of a rather wide experience with splenectomy, the operation came to be reserved for the young person with early Banti's syndrome

It is obvious that these two surgical procedures, omentopexy and splenec-

tomy, dominated therapy prior to 1940. Their general lack of success and an increasing awareness of the clinical importance of hemorrhage from varices stimulated the development of other measures directed more specifically at the control of bleeding. In 1926, Flerow advocated ligation of the left gastric artery, the right and left gastro-epiploic arteries, and the inferior mesenteric vein. A few years later, Walters and his associates re-



hemorrhage

ported some success following ligation of the left gastric veins In 1933, Holman expressed the belief that bleeding could be controlled by ligation of the splenic artery and implantation of the spleen itself into the abdominal wall In a recent article, Gerbode and Holman reported that two patients upon whom this operation had been done had not experienced a recurrence of hemorrhage over a period of 12 years In 1939, Crafoord and Freckner demonstrated that the varices could be injected transesophageally with a sclerosing solution. This technique was practiced enthusiastically by Moersch in this country, but has not been widely accepted. Because of the

scarcity of good results attributable to these procedures, they have generally been abandoned.

The modern concept of the treatment of portal hypertension was developed in the Spleen Clinic at the Presbyterian Hospital in New York City. In 1934, the Spleen Clinic under the direction of Dr. Allen O. Whipple was organized. Members of its staff came from the Departments of Internal Medicine, Surgery, Hematology and Pathology. In 1936, Rousselot, a member of this group, published his first report. Discussing "The Portal Vascular Bed", he wrote, "Dr. J L. Caughey of our Medical Staff recently has been making some interesting studies on venous pressure in the splenic vein as seen at the operating table. ... These have shown a definite rise in certain cases of hepatosplenomegaly ... Possibly herein lies a clue to the solution of the cause of this confusing syndrome." During the next six years this group studied many patients with esophagogastric hemorrhage and became convinced that the etiological factor involved was portal hypertension. In 1942, therefore, Rousselot* and Whipple undertook direct decompression of the hypertensive portal bed by anastomoses between "smaller branches of the mesenteric veins" and "smaller radicles of the caval system" These uniformly failed In 1945, Blakemore and Lord reported on their non-suture method of blood vessel anastomosis. So encouraged was Blakemore by his results in arterial anastomosis that he was led to try the method for the establishment of portacaval shunts. Although non-suture methods have now been abandoned in favor of direct suture technique, they established large vessel decompression of the hypertensive portal bed as a feasible procedure As the direct results of the efforts of Whipple, Rousselot and Blakemore, surgeons in this country and abroad have been stimulated to undertake again the treatment of those unfortunate patients apparently doomed to die from esophagogastric hemorrhage before the disease has advanced too far In the following paragraphs are discussed the current methods of portal decompression.

Portal Decompression by Veno-Venous Shunts

Only the splenic and portal veins are of sufficient size to be important in portal bed decompression Shunts performed between the portal vein and the inferior vena cava are commonly referred to as portacaval (fig 2A and B), while those between the splenic vein and the left renal vein are termed splenorenal (fig 3A and B). Rarely other vessels such as the superior mesenteric and right renal vein have been used to form a portacaval anastomosis. Such shunts almost uniformly fail to remain patent. Experience with portacaval and with splenorenal shunts is now sufficiently extensive to permit their evaluation in some detail

* Personal communication

A

B

B

Portacaval Versus Splenorenal Shunts

In patients in whom both the splenic and portal veins are equally suitable for anastomotic purposes, there is no generally preferred procedure. Blakemore favors the portal vein because it is large, especially in patients with cirrhosis. Here its generous caliber and healthy walls permit making a shunt between it and the inferior vena cava which decompresses the portal bed effectively. Postoperatively varices can no longer be seen roentgenographically, the spleen regularly shrinks in size, and follow-up studies indicate that most patients had no further hemorrhage (Blakemore). At The New York Hospital, the portacaval shunt has been employed in 24 patients with intrahepatic block. To date, the results have been satisfactory. The following is typical of patients in whom a portacaval shunt is most useful

Case No 2, S L Female Age 47 NYH #589 381. S. L, a 47-year-old housewife who, over the years had drunk large quantities of whiskey and sherry, was admitted following her second episode of gastrointestinal hemorrhage. Her first had occurred two years before at which time she was told by her physician that she had cirrhosis of the liver

Examination revealed an obese woman in apparent good health who complained of dizziness and dyspnea on the slightest exertion. A firm liver edge was palpable three centimeters below the right costal margin. Laboratory studies revealed a hemoglobin of 9 grams and RBC of 2 9 million. Other blood chemical determinations were within normal limits. An esophagogram revealed large varices. At operation, her liver was cirrhotic, the portal pressure was 42 cm of saline. This fell to 27 cm after establishing the end-to-side portacaval shunt. Her postoperative course was uncomplicated, and she has not bled during the first year after operation. Five months after operation, roentgen examination of the esophagus revealed great diminution in the size of the varices

Rousselot and Linton prefer the splenorenal shunt. They insist that this type of shunt offers quite as great protection against further hemorrhage as does the portacaval. In figure 5A and B are reproduced the preoperative and six-month postoperative esophagograms of a patient (J. K., NYH #603 401) in whom splenic venous pressure was reduced from 46 to 23 cm of saline by an end to side splenorenal shunt. The extensive varices present preoperatively cannot be demonstrated in the postoperative film.

Both these authorities believe that removal of the spleen is in itself an important aspect of the treatment of patients with portal hypertension. Not only does splenectomy reduce the amount of blood entering the portal bed, but in addition excessive blood destruction is prevented. Linton reserves the portacaval shunt for persons whose spleen is small and whose hemogram is normal. If at operation either the portal or the splenic vein is unsuitable for constructing a shunt, the choice is automatic. For instance, if a patient has continued to bleed after a splenectomy and a healthy

portal vein can be found, a portacaval shunt is indicated. If, as frequently occurs in patients with extrahepatic block, the portal vein is partially or wholly unavailable and a splenic vein is normal, a splenorenal shunt should be performed.

In considering these shunts, one unsolved problem is whether or not to preserve a certain proportion of portal blood flow through the liver. This is of no particular concern in the splenorenal shunt where portal blood flow is undisturbed save that its head of pressure is conspicuously lowered. A portacaval shunt, on the other hand, usually can be performed in either the lateral or end-to-side position. In the lateral position, a portion of portal blood continues to flow through the liver; in the end-to-end position *portal blood is completely diverted from the liver* A number of experimental observations are advanced by those who favor the lateral type of anastomosis. It has been established by numerous investigators (Rous, Larrimore, Mann, Fishback, Higgins, Markowitz) that in the rabbit, cat, and dog the liver will not regenerate if portal vein flow is interrupted In laboratory animals with an end-to-side portacaval shunt an abnormality in amino acid levels in the peripheral blood occurs when they are given by stomach tube (Harper and Gardner). Preshaw and his associates have shown in the dog that bromsulfalein retention is appreciably greater after an end-to-side portacaval shunt (fig. 2A), then after a lateral anastomosis (fig 2B). Dogs with the end-to-side portacaval shunt are far more susceptible to carbon tetrachloride poisoning than are those with the lateral shunt Whether these observations can be applied validly to man has not been established Those who do not believe that these facts apply in man advance arguments against the lateral anastomosis which cannot be disregarded Hypertrophy of the caudate lobe may make a lateral anastomosis hazardous In dogs even a generous lateral anastomosis shows a great tendency to shrink in size postoperatively to little more than a few millimeters or so in diameter Furthermore, it can be argued with some justification that in cirrhosis, high intrahepatic resistance will in itself prevent portal blood from flowing through the liver after portal pressure has been reduced by a lateral portacaval shunt Actually then, after such a shunt has been opened all portal blood and perhaps even an increment of hepatic blood may escape directly into the vena cava

No matter what the final decision to this complex problem may be, there is evidence today indicating that the lateral anastomosis is less efficient in lowering portal pressure For instance, Julian and Fildes report the following falls in centimeters of saline in three patients with lateral anastomoses: 50 to 40, 45 8 to 42 4, and 39 6 to 30 In Blakemore's series patients with the shunt in the end-to-side position manifested decreases of the following order: 37 to 23, 46 to 19, 56 to 22, and 45 to 24 centimeters

of saline. In The New York Hospital series where end-to-side anastomoses have generally been performed, falls in pressure from 35 to 45 centimeters of saline down to 18 to 23 centimeters of saline have been obtained frequently. Currently, it is the consensus that differentials in liver function or in well-being of the patient cannot be demonstrated when the end-to-side portacaval shunt is compared with the lateral portacaval anastomosis. Until specific evidence in favor of the lateral type of anastomosis is produced, the end-to-side is probably preferable.

In Blakemore and Lord's original paper, they recorded five patients in whom splenorenal shunts were performed In each instance, both the spleen and kidney were removed and the anastomosis constructed between the end of the splenic and end of the renal vein (fig. 3B). They concluded at the time that an end-to-side anastomosis (fig 3A) rather than one in the end-to-end position would obviate the necessity for sacrificing one kidney. As Linton was developing his experience with the splenorenal shunt, he objected to the loss of *one* kidney in a group of patients whose clinical status was at best precarious Furthermore, attention has repeatedly been called to the fact that patients who have cirrhosis of the liver may have significant degrees of renal damage (Baxter and Lichtman). For this reason, Linton insisted that the splenorenal shunt be performed in the end-to-side position, thereby preserving the left kidney As experience has been gained with this type of decompression, the end-to-side position of the shunt has been accepted as the best procedure In the course of such an operation, the blood flow in the renal vein may be interrupted temporarily without damage to the kidney by occluding the renal artery Normal renal function has been demonstrated after many such operations An alternate method, which is probably more widely favored today, involves the use of a special clamp (fig 4A and B) which permits isolation of a segment of vein while allowing renal venous flow to continue only partially impeded Although a final evaluation of these two methods is not yet possible, temporary occlusion of the renal artery has in its favor the fact that a somewhat larger shunt can be obtained while its disadvantage rests in the temporary occlusion of all renal blood flow This can hardly be regarded without at least some concern because of the supposed direct relationships between renal anoxia and the development of the shock kidney In employing specially devised clamps renal flow is not significantly compromised, but there is some question as to the effective size of the anastomosis which can be obtained

As a result of his recent experiences with splenorenal shunts, Rousselot has developed a technique which uses a graft from the superficial femoral vein This is interposed between the end of the splenic vein and the side of the renal (fig 6A). By the use of this type of free graft, Rousselot be-

lieves that the shunt can be constructed more easily and upon its completion can be relied upon to function more effectively than where the graft is not employed. If splenectomy is ultimately adopted as part of all operations for portal hypertension, this technical addition may prove useful.

Ample evidence is available today proving that if a satisfactory shunt can be constructed, an impressive fall in portal pressure follows. Although this may not be to normal levels, it is still low enough to protect the patient

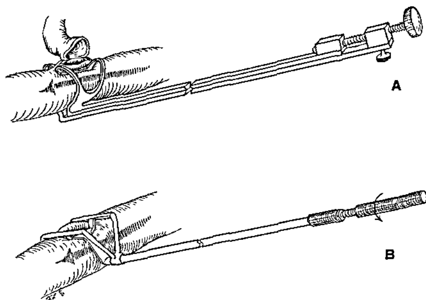


FIG 4 TWO VARIETIES OF CLAMP ESSENTIAL FOR THE CONSTRUCTION OF

be exposed

from repeated massive hemorrhages The question of how long and how successfully these shunts maintain a reduction in portal pressure, has been asked repeatedly. At the present time, a reasonably complete answer to this question can be given. That the anastomoses do remain open and do protect patients from further hemorrhage is reflected strikingly in Blakemore's recent report. Among a total of 38 portacaval and 38 splenorenal shunts, 12 patients died during the follow-up period Only 5 suffered from recurrent hematemesis and in these the shunts were shown to have closed Only 2 patients with open shunts bled postoperatively. Linton has reported a series of 61 patients, 46 with a splenorenal shunt and 15 with a

portavacal, without a death from recurrent hemorrhage. Each reported series contains a few patients in whom either a satisfactory shunt could not be obtained or the shunt became closed. Nearly all are known to have suffered recurrent hematemesis. At present, therefore, it is justifiable to conclude that if a satisfactory fall in portal pressure is obtained following either a portacaval or splenorenal shunt, most patients can expect protection against further hemorrhage for at least a limited period of time. Final evaluation of portal decompression must await studies on patients followed over a longer period.

Recently Blakemore has called attention to a small group of patients with extrahepatic portal hypertension who have bled repeatedly following splenectomy alone. When both the portal and splenic veins were uniformly unfit for shunting purposes, Blakemore, Rousselot, Reynolds, and Southwick have demonstrated the practicability of securing decompression by means of a free vein graft In several patients, they inserted such a graft between partially compromised portal or splenic veins and the inferior vena cava (fig. 6B).

Non-Shunting Procedures for the Relief of Portal Hypertension

Interestingly enough, the establishment of venous shunts as a sound surgical procedure has produced a number of other operations aimed directly at removing the site of hemorrhage The proponents of these procedures readily admit that where a shunt can be constructed satisfactorily, it seems to be the most logical form of therapy for the patient with repeated esophagogastric hemorrhage from varices. A shunt may not always be possible or may fail. To complete the picture of surgical therapy for portal hypertension, these additional procedures are considered.

The most important of these operations, esophagogastrectomy, was introduced by Phemister in 1947 Encouraged by his success in treating esophagogastric cancer in this manner, he proposed the operation as a life-saving measure for patients with bleeding from esophagogastric varices. Its benefits, of course, are derived directly from the removal of the site of hemorrhage In his discussion of Phemister's original report Wangensteen proposed an additional concept· namely, acid-peptic erosion of the esophagus as the cause of bleeding from varices. This approach to esophagogastric hemorrhage has been widely elaborated by Baronofsky. Wangensteen later indicated that these erosions might be prevented were a total gastrectomy performed. Several other surgeons have been attracted to this idea of local resections, and recently Womack, Schafer and Kittle, Gray and Whitesell have advocated esophagogastrectomy together with splenectomy. The latter authors have gone one step further by adding vagotomy in order to eliminate the cephalic phase of acid gastric secretion The following case

is an example of one requiring direct removal of the area of stomach and esophagus from which bleeding presumably occurred.

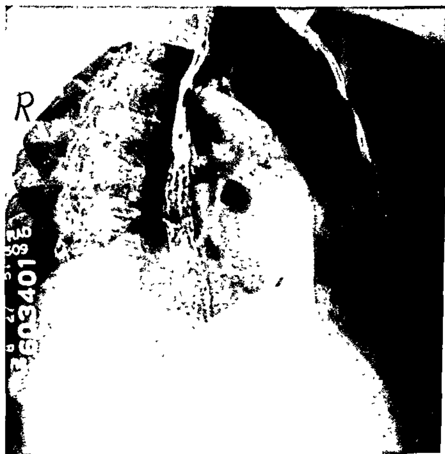


FIG 5A AND B J K NYH #603 401 PREOPERATIVE AND POSTOPERATIVE ESOPHAGOGRAMS

Case No 3 Esophagogastrectomy for Bleeding Esophageal Varices D R NYH

This amounted to only a few centimeters of saline. It did not seem justifiable to attempt any further procedure at this time, and the abdomen was closed.

During the next 18 months, she had two additional episodes of severe hemorrhage. She was again admitted and at operation the portal venogram shown in figure 7 was obtained The granulomatous mass had become sclerotic, and the portal vein was unavailable for decompression. The splenorenal shunt had failed An esophagogas-

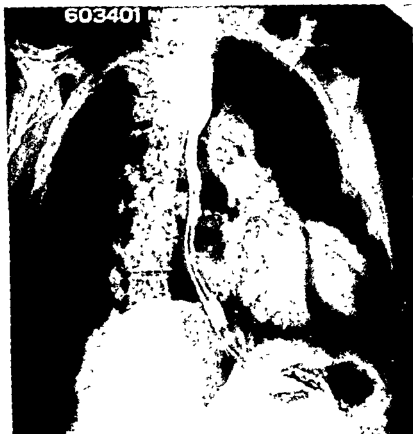
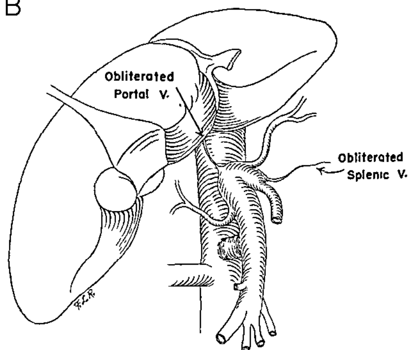


Fig 5B

trectomy was tried to prevent further hemorrhage Both vagi were divided The continuity of the upper enteric canal was reestablished by a Roux "Y" esophagojejunostomy The proximal end of the stomach was closed Anastomosis was effected between the esophagus and jejunum rather than between the esophagus and stomach in the belief that erosion of the lower esophagus by acid-peptic gastric contents might be avoided One-half of the stomach was retained, hoping thereby to prevent a post-total-gastrectomy nutritional disaster A pyloroplasty was performed to avoid difficulty in gastric emptying from bilateral vagotomy It is too early in her follow-up period to evaluate results She left the hospital on the 23rd postoperative day in apparent good health The details of this extensive procedure are diagrammatically

A

B



portrayed in figure 8. An esophagogram performed two months after operation failed to visualize varices.

In addition to one form or another of partial esophagogastrectomy, other procedures have been suggested. In 1947, Som and Garlock reported two



Fig. 7. Case 4. D. D. NYH [illegible]

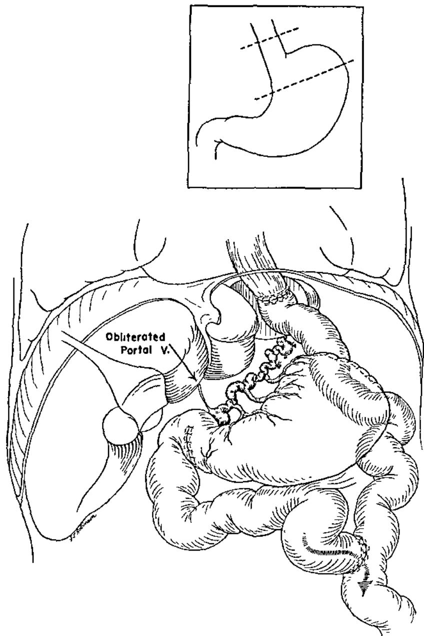


FIG 8 CASE 4 D R NYH #576 533 ESOPHAGOGASTRECTOMY FOR ESOPHAGOGASTRIC HEMORRHAGE

cases in which they packed the upper thoracic mediastinum with gauze. They expressed the hope that the granulations forming in response to foreign material would develop an extensive periesophageal plexus capable of reducing the pressure in the varices. Garlock and Som recently reviewed their experiences with this procedure in 8 cases, and they still believe that packing produces a periesophageal collateral circulation. They concluded that it will require further experience before final evaluation of their method can be reached.

Because the splenic artery contributes a considerable portion of blood to the portal bed, Holman, in 1933, suggested its ligation as a method of decreasing portal blood flow. In addition, he advocated implantation of the spleen into the abdominal wall as a method for the promotion of venous drainage. Recently Blain and Blain have advocated splenic artery ligation alone as a procedure useful in those patients who could not under any circumstances be expected to survive any procedure of great magnitude.

The most recently suggested form of therapy for portal hypertension is ligation of the hepatic artery as advocated by Rienhoff in 1951. This procedure is based upon Herrick's work published in 1907. Herrick believed he had demonstrated the presence of direct arteriovenous communications between the hepatic artery and the portal vein in patients with cirrhosis of the liver. Although McIndoe was unable to confirm Herrick's findings, there is evidence that hepatic arterial blood pressure may contribute to the portal hypertension in patients with cirrhosis. Because portacaval and splenorenal shunts are long and difficult surgical procedures, there is every incentive to devise a simpler form of treatment. When the construction of one of these shunts is compared with ligation of the hepatic artery it is obvious that the latter is far easier and less time-consuming. Should hepatic artery ligation prove effective, it would supercede direct portal decompression At the present time, too few cases have been operated upon to permit even a preliminary evaluation of this procedure. Rienhoff recently reported 13 patients who have survived this operation and presumably been improved Berman in an article on experimental ligation of the hepatic artery refers briefly to several patients who were benefited by this procedure. Another form of therapy that has been advocated is that of Crile who maintains that the varices may be regarded somewhat as are hemorrhoids In 1950, Crile demonstrated that the varices could be resected trans-esophageally In the same year, Allison suggested periesophageal devascularization as a procedure of potential usefulness.

The Selection for Operation of Patients with Portal Hypertension

Earlier in this chapter, evidence was advanced indicating that in man hypertension develops in the portal venous system in response to partial or complete obstruction to portal blood flow. Two types of obstruction are

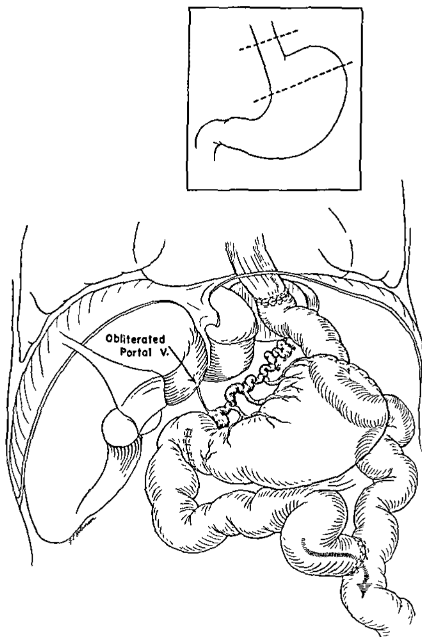


FIG 8. CASE 4 D R. NYH #576 533 ESOPHAGOGASTRECTOMY FOR ESOPHAGOGASTRIC HEMORRHAGE

In this patient, portal decompression was impossible The lower third of the esophagus and upper third of the stomach were removed Reconstruction of the

suitable candidates for portal decompression. This is to be expected since fluid retention and jaundice are the two main clinical manifestations of liver failure. Lichtman has pointed out that hematemesis immediately precedes jaundice and ascites often enough to suggest that these symptoms are associated with developing liver decompensation. This has, indeed, been the experience in The New York Hospital Liver Clinic where prognosis for the patient with cirrhosis is considered grave once one or more of this triad of symptoms appears.

CIRRHOSIS—HEMORRHAGE WITH ASCITES AND/OR JAUNDICE

Decision as to whether to recommend operation for patients falling into this general category is difficult. Once fluid retention or jaundice has appeared, the prognosis is poor. If a patient, however, improves on a conservative regime so that he is no longer jaundiced and is able once again to handle at least a minimum fluid load, he may withstand operation amazingly well. The patient who fails to respond to a supportive regime and who continues to accumulate fluid despite rest, high protein diet, and the usual methods of fluid control, i e., mercurials and low salt diet, is a poor candidate for operation. Such a patient may go into hepatic coma with anuria, or may continue to pour ascitic fluid into the peritoneal cavity, thus depleting the protein reserve as well as the sodium stores of the body. In such patients, the operative mortality is so high and the chance of hepatic failure so great that surgical intervention should not be undertaken

Under certain extenuating circumstances, however, an effort to decompress the portal bed may become justified almost as an emergency procedure Where it appears obvious that a patient will die from hemorrhage or at least that he cannot be sustained forever by transfusion, what reasonable course can be followed? Today there is no unanimity of opinion about management Although emergency shunts have been performed successfully, there is little reason to hope that success under such circumstances will be the rule Some have suggested such emergency procedures as transthoracic ligation of the esophageal bleeding points and even esophagogastrectomy Both of these operations are almost as formidable as a shunt and hardly seem to be the answer to this difficult problem The following case is considered an example of the many difficulties which confront those assuming the responsibility for the care of patients in this category:

Case No 4 M A Female Age 46 NYH #601 270 Cirrhosis of the Liver Esophageal varices [illegible] ... l protein ... ousewife

The initial manifestations of her hepatic disease appeared five years before admission These were intermittent attacks of vague upper abdominal pain associated with nausea and vomiting From time to time during the next several years, she noted

commonly encountered; those within and those without the liver. One of the well-recognized manifestations of increased portal pressure is esophagogastric varices. That hemorrhage from these may seriously threaten life in a patient with either form of portal obstruction has been recognized for many years. Recently, methods have been developed whereby acute hemorrhage can be controlled, and the incidence of recurrent episodes of bleeding materially reduced. Concurrent with the development of these therapeutic measures there has appeared a relatively new clinical problem—namely, which patients with portal hypertension should be selected for operation?

In patients whose esophagogastric varices result from extrahepatic block, the decision to recommend operation is relatively uncomplicated. Generally, these are young persons in good health They can be expected to stand operation well. In the majority of instances, portal decompression can be effected by splenectomy and splenorenal shunt. Rarely, a vein graft will be required to provide for the transfer of blood from the portal to the systemic circulation Where neither of these methods is available, esophagogastrectomy has been employed successfully.

When, however, hemorrhage from esophagogastric varices occurs in the patient with cirrhosis, the problems involved in the selection of those suitable for a portacaval shunt are complex. Non-surgical therapy has little to offer in the control of hemorrhage The mortality rate in patients with cirrhosis who have had one hematemesis is high However, only one-fifth of the patients with cirrhosis in Patek's series died of hemorrhage and four-fifths as the result of liver failure In any patient with cirrhosis who has had one hemorrhage, the decision whether to recommend operation depends upon whether esophagogastric hemorrhage or hepatic failure is the greater hazard to life

In reaching this decision, an accurate evaluation of the functional reserve of the liver is important. The large number of liver function tests which have been devised for this purpose is testimony of the many difficulties involved. There is no single test of liver function which correlates consistently with the clinical course of the patient Although a group of such tests gives a better survey of the functional reserve of the liver, they cannot provide the final answer as to whether or not the patient should be subjected to operation.

Linton has suggested a clinical classification that provides a workable basis for deciding which patients with cirrhosis should be operated upon It serves to clarify some of the current concepts on the indications for portal decompression With a few modifications it is outlined below:

CIRRHOSIS—HEMORRHAGE WITHOUT ASCITES OR JAUNDICE

Of the patients with cirrhosis who have bled from esophagogastric varices, those who have never developed ascites or jaundice are considered the most

CIRRHOSIS WITHOUT HEMORRHAGE BUT WITH VARICES

Opinion is divided about operation for the patient with cirrhosis and varices who has never bled. For instance, Julian and Fildes accept the demonstration of varices as indication for operation provided there is reasonable hepatic reserve. Linton, on the other hand, does not believe operation should be considered merely because varices have been demonstrated. At The New York Hospital, mere demonstration of varices has not been considered an indication for portal decompression. This attitude has been taken with full recognition of the seriousness of an episode of hemorrhage in the life history of patients with cirrhosis. As more experience is acquired, it may prove desirable to attempt to forestall such an episode by a prophylactic portacaval shunt.

CIRRHOSIS WITH UNCONTROLLED ASCITES

When portal decompression was first introduced, many patients with cirrhosis and massive ascites were subjected to this operation. It was hoped that troublesome re-accumulation of ascites could be prevented by reducing portal pressure. Not only was the ascites unaffected, but in this group the mortality was found to be prohibitively high. Currently, the operation is not performed with ascites as the primary indication.

Liver Function Tests as an Aid in Selection of Patients for Portal Decompression

In the section dealing with the selection of patients for portal decompression, any evaluation of the currently performed liver function tests was purposely omitted. While significant advances have been made in estimating liver function by means of biochemical tests, these have proved disappointing as a basis for the selection of those patients with esophagogastric hemorrhage who should be subjected to operation. In our experience, levels of serum albumin and serum bilirubin have been most useful in reaching a decision. Others accepting the responsibility for the care of these patients have found a valid correlation between high bromsulfalein retention and postoperative morbidity and mortality. As in any form of liver disease, prolonged prothrombin time uncorrectable by the administration of Vitamin K is a deterrent to operative intervention. One value accorded particular attention in any major surgical problem is the level of serum albumin. It is generally conceded that where this is 3 grams per 100 milliliters or below, operation will be poorly tolerated. It has been the general policy of this clinic, therefore, to withhold portal decompression in patients whose serum albumin is significantly below this level. In addition, operation is not advised where serum bilirubin rises much above 4 milli-

dependent edema and had several bouts of epistaxis During this period, her alcoholic intake was high, and her diet inadequate Eight months before admission she was acutely ill for several weeks, ran a spiking fever and later became jaundiced A few months later, ascites made its appearance This required several paracenteses During the month before admission, she was treated in another hospital where her fever was persistent, *anemia was marked, and she was occasionally semi-comatose*

On admission, she was frail, chronically ill, febrile and wasted The striking features on physical examination were spider angiomata, dilated abdominothoracic veins and ascites Of the numerous laboratory studies which were performed, the following seemed most significant serum bilirubin 25 milligrams per cent, alkaline phosphatase 3 8 Bodansky units, hemoglobin 9 8 grams per cent; RBC 3 1 million, stool positive for blood Esophageal varices were demonstrated roentgenographically. Prothrombin time was prolonged to 18 7 seconds compared with a control of 14 3 Only by repeated transfusions was it possible to maintain a serum albumin level between 3 4 and 4 0 grams per cent, and her hemoglobin and red count at reasonable levels Although it was accepted that the risk was great, it was decided to subject this patient to operation in an effort to control persistent bleeding An end-to-side anastomosis was easily accomplished and the procedure was well tolerated After completion of the shunt the pressure fell from 40 to 15 cm of saline For the first few postoperative days, she did reasonably well, but then passed into a phase of rapid accumulation of ascitic fluid, low serum sodium, low serum albumin, and oliguria In spite of vigorous supportive therapy, the abnormalities in her serum levels could not be corrected Coma, arterial hypotension, and oliguria made their appearance, and she died on the *eleventh day after operation*

A post-mortem examination limited to the abdominal cavity revealed cirrhosis (Laennec type) of the liver, extensive esophagogastric varices, and a portacaval shunt occluded by thrombus There was ischemic necrosis of 6 to 7 millimeters of portal vein immediately adjacent to the site of anastomosis

Comment. In retrospect, this woman should probably not have been subjected to operation. She is an example, however, of that group of patients who are bleeding, who are jaundiced, who have ascites, and in whom operation apparently offers the only chance of survival Apparently, she was in the terminal phase of her disease and unfortunately, a technically perfect shunt became occluded by a small thrombus.

Three groups of patients are not considered candidates for portal decompression. These are·

CIRRHOSIS WITHOUT HEMORRHAGE OR VARICES

Patients in whom the diagnosis of cirrhosis of the liver can readily be established but who have never bled and in whom esophagogastric varices cannot be demonstrated roentgenographically should probably not be operated on Failure to identify varices is considered good evidence that significant degrees of portal hypertension do not exist Since there is little if any reason to believe that a portacaval or splenorenal shunt improves liver function, there seems at present no reason to suggest operation for the patient with uncomplicated cirrhosis

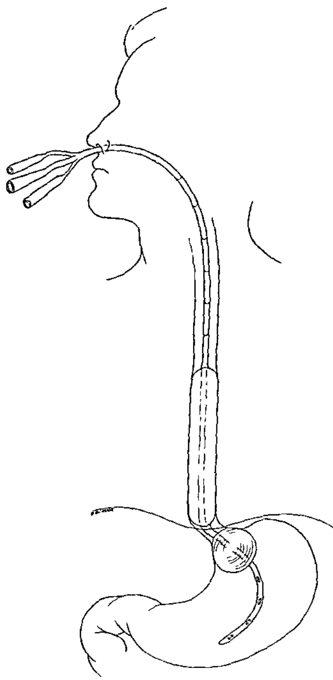
grams per 100 milliliters, and a significantly prolonged prothrombin time cannot be corrected by the parenteral administration of Vitamin K.

Balloon Tamponade in the Emergency Treatment of Hemorrhage from Esophagogastric Varices

As long ago as 1900, the idea of controlling hemorrhage from esophageal varices by a rubber balloon was proposed (Preble). Since then, a number of individuals have reported varying degrees of success with balloons of one type or another. It was not, however, until 1946 that Sengstaken and Blakemore developed a balloon which proved satisfactory. The design of this instrument and the position it occupies when inflated in the stomach and lower esophagus are depicted diagrammatically in figure 9. This balloon has several advantages over the older types. It maintains its position in the esophagus instead of working into the stomach with peristaltic waves. The separate gastric balloon is useful in anchoring the esophageal section snugly against the cardia of the stomach and for actual tamponade of gastric varices. In addition, the free tube to the stomach permits gastric suction while the balloon is still inflated. This diminishes nausea and retching, and allows the physican to determine whether the bleeding is under control. At 24- to 48-hour intervals, the balloon can be deflated but left in place. In this fashion the patient may be maintained with relative safety during the preparation for portal decompression. Once the bleeding has stopped, the patient can be given fluids and nourishment through the tube. Esophageal tamponade should be discontinued as soon as possible after bleeding has ceased. Ulceration of the nose, throat and esophagus can occur when the balloon is left in place for too long a period of time.

BIBLIOGRAPHY

1. Ahrens, E. H., Jr., Payne, M. A., Kunkel, H. G., Eisenmenger, W. J. and Blondheim, S. H.: Primary biliary cirrhosis. Medicine, 29: 299, 1950
2. Allison, Phillip Rowlands. Discussion of H. K. Gray and F. B. Whitesell. Hemorrhage from esophageal varices. Surgical management. Ann. Surg., 132: 808, 1950.
3. Andrus, W. deW. and Holman, C. W.: Splenectomy in various blood disorders. Ann. Surg., 109: 64, 1939.
4. Aschoff. Quoted by J. McMichael: The pathology of hepatolienal fibrosis. J. Path. & Bact., 39: 481, 1934.
5. Banti, Guido: Dell anemia splenica. Arch. d. Scuola d'anat. Patol., 2: 1883.
6. Banti, Guido: La splénomégalie avec cirrhose du foie. Sem. Méd., 14: 318, 1894.
7. Banti, Guido. Splenomegalie mit Lebercirrhose. Beitr. z. path. Anatomie u. z. allg. Pathologie, 24: 21, 1898.
8. Banti, Guido: Ueber Morbus Banti. Folia haematol., 10: 33, 1910.
9. Baronofsky, Ivan D. Portal hypertension. With special reference to the acid-peptic factor in the causation of hemorrhage and extensive gastric resection. Surgery, 25: 135, 1949.



30. CAMERON, G. R. AND DE SARAM, G. S W.: A method for permanently dissociating the spleen from the portal circulation (the "marsupialised" spleen) and its use in the study of experimental liver cirrhosis. J. Path. & Bact , 48: 41, 1939.
31. CATES, HORACE B.: Surgical treatment for cirrhosis. Prognosis subsequent to omentopexy. Arch Int. Med , 71: 183, 1943.
32. CAUCHOIS, ANDRÉ· Splénomegalie chronique d'origine pyelthrombosiquem Paris, G Steinheil, 1908.
33 CAUDHEY, J. L.: Quoted by L. M. ROUSSELOT: The role of congestion (portal hypertension) in so-called Banti's syndrome. A clinical and pathologic study of thirty-one cases with late results following splenectomy. J. A. M. A , 107: 1788, 1936
34. CHIARI, H.: Uber die selbstandige Phlebitis obliternas der Hauptstamme der Venae hepaticae als Todesursache. Beitr z. path Anat. u. z. allg Path , 26: 1-18, 1899
35 CHILD, CHARLES G , III., MCCLURE, ROY D , JR AND HAYS, DANIEL M.: Studies of the hepatic circulation in the *Macaca Mulatta* monkey and in man Surg Forum, Thirty-seventh Clinical Congress Am Coll. Surgeons, San Francisco, Calif., November, 1951, W. B Saunders Co
36 CHILD, CHARLES G , III, MILNES, CHARLES G , HOLSWADE, ROGER F. AND GORE ARTHUR L.: Sudden and complete occlusion of the portal vein in the *Macaca Mulatta* Monkey. Ann Surg , 132: 475, 1950
37 CHILD, CHARLES G , III, HOLSWADE, GEORGE R , MCCLURE, ROY D , JR , GORE, ARTHUR L AND O'NEILL, EARL A · Pancreaticoduodenectomy with resection of the portal vein in the Macaca Mulatta monkey and in man Surg , Gyn & Ob , 94: 31, 1952.
38 CRAFOORD, CLARENCE AND FRENKNER, PAUL: New surgical treatment of varicose veins of the oesophagus. Acta oto-laryng , 27: 422, 1939
39 CRILE, GEORGE, JR Transesophageal ligation of bleeding esophageal varicose A preliminary report of seven cases Arch Surg., 61: 654, 1950
40 DAVIDSON, C S , GIBBONS, R B. AND FALOON, W W.: Systemic and portal venous pressures in cirrhosis of liver J. Lab and Clin Med., 35: 181-187, 1950.
41 DOCK, WILLIAM The role of increased hepatic and arterial flow in the portal hypertension of cirrhosis Trans. Ass Am Phys , 57: 302, 1942
42 DOUGLASS, BRUCE E AND SNELL, ALBERT M : Portal cirrhosis an analysis of 444 cases with notes on modern methods of treatment Gastroent , 15: 407, 1950
43 ECK, NIKOLAI VLADIMIROVICH· The ligature of the portal vein Voyenno-med J , 130: 1, 1877
44 EDENS Ueber Milzvenenthrombose, Pfortaderthrombose und Bantische Krankheit Mitt a d. Grenzgeb. d Med u Chir , 18: 59, 1908
45 EDWARDS, E A Functional anatomy of the porta systemic communications. Arch Int Med , 88. 137-154, 1951
46 ELMAN, ROBERT AND COLE, WARREN H . Hemorrhage and shock as causes of death following acute portal obstruction Arch Surg , 28: 1166, 1934.
47 EPPINGER, H Die hepatolienalen Erkrankungen, pp 384-493 Berlin, Julius Springer, 1920
48 FAUVEL. Bull soc méd. d'obs , 1858
49 FISHBACK, FREDERICK C A morphologic study of regeneration of the liver after partial removal Arch Path , 7: 955, 1929

10. BAUDOUIN, M.· La ligature de l'artère hépatique. Ann Intern de chir. gastro-intest., 3: 39, 1909.
11. BAXTER, J. H. AND ASHWORTH, C. T.: Renal lesions in portal cirrhosis Arch Path , 41: 476, 1946
12. BAYLISS, W. M AND STARLING, ERNEST H.: Observations on venous pressures and their relationship to capillary pressures J. Physiol., 16: 159, 1894
13. BEAN, W. B , FRANKLIN, M., EMBICK, J F. AND DAUM, K : Absorption studies using portal anastomotic veins J. Clin Inv , 30: 263-273, 1951
14. BERMAN, JACOB K.. Discussion of J H. GRINDLAY, F C. MANN AND J. L BOLLMAN. Effect of occlusion of the arterial blood supply in the normal liver An experimental study Arch. Surg , 62: 810, 1951
15 BERMAN, JACOB K , MULLER, LULLUS P., FISCH, CHARLES AND MARTZ, WILLIAM Ligation of the hepatic and splenic arteries in a patient with atrophic cirrhosis of the liver Arch Surg , 62: 623, 1951
16 BERNARD, CLAUDE. Leçons sur le Diabète et la Glycogenèse Animale VIII 576 pp Paris, J. B Baillière & fils, 1877
17. BILLINGS, F. T , JR AND DEPREE, H S Diagnosis of portal vein obstruction, studies of intestinal absorption of glucose using abdominal collateral veins Bull Johns Hopkins Hosp , 86: 183–199, 1949
18. BLAIN, ALEXANDER W AND BLAIN, ALEXANDER, III Ligation of the splenic artery, operation of choice in selected cases of portal hypertension and Banti's syndrome Ann Surg , 131: 92, 1950
19 BLAKEMORE, ARTHUR H Portacaval shunts for portal hypertension Follow up *results in cases of cirrhosis of the liver* J A M A , *145: 1335,* 1951
20 BLAKEMORE, ARTHUR H AND LORD, JERE W , JR : A nonsuture method of blood vessel anastomosis Experimental and clinical study J. A M A , 127: 685 & 748, 1948
21 BLAKEMORE, ARTHUR H AND LORD, JERE W , JR The technique of using vitallium tubes in establishing portacaval shunts for portal hypertension Ann Surg , 122: 476, 1945
22 BLAKEMORE, ARTHUR H AND FITZPATRICK, HUGH F The surgical management of the post-splenectomy bleeder with extra hepatic portal hypertension Ann Surg , 134: 430, 1951
23. BLONDHEIM, S H AND KUNKEL, G H *Portal blood in collateral veins of* patients with cirrhosis, acetylation by intestine Soc Exp Biol and Med , 73: 38, 1950
24. BLOOM, H J G Venous hums in hepatic cirrhosis Brit Heart J , 12: 343–350, 1950
25. BOGORAS, N. Ueber die Ueberpflanzung der Vena mesenterica superior in die [illegible]

2[illegible]
circulation Liver Injury Trans , 7th Conf , Macy Foundation, 21, 1948
27 BOYCE, F F *The Rôle of the Liver in Surgery (Chap X) Springfield, Ill ,* Charles C Thomas, 1941
28 BRANDES, W W The effect of mechanical constriction of the hepatic veins with special reference to the coagulation of blood Arch Int Med , 44· 676, 1929
29. BROWN, LANGDON, W. "Pyelephlebitis " St Barth Hosp Rep , 37: 53, 1901

72 LARRABEE, RALPH C. Chronic congestive splenomegaly and its relationship to Banti's disease Am. J. Med Sc, 188: 745, 1934.
73 LAUTENBACH, B. F.: On a new function of the liver Phila. Med Times, 7: 387, 1877.
74. LAZZARI, JOHN H. AND RACH, FRANK J · Hepar lobatum with portal hypertension successfully treated by portacaval anastomosis Arch. Surg, 62: 295, 1951.
75. LENOIR Quoted by ENDERLEN, HOTZ AND MAGNUS-ALSLEBEN: Die Pathologie und Therapie des Pfortaderverschlusses Experimentelle Untersuchungen ueber die Eck'sche Fistel Ztschr. f d ges. exper. Med, 3: 262-263, 1914.
76 LEVY, L. K. AND BURCH, G. E Studies on venous pressure in hepatic cirrhosis Ann. Int. Med, 29: 274-277, 1948
77. LICHTMAN, S S. Diseases of the Liver, Gallbladder and Bile Ducts. Ed 2 Phila, Lea & Febiger, 1949
78 LICHTMAN, S S AND SOHVAL, A R: Clinical disorders with associated hepatic and renal manifestations, with especial reference to the so-called "hepatorenal syndrome" Am. J Dig. Dis, 4: 26, 1937
79 LINTON, ROBERT R The selection of patients for portacaval shunts. With a summary of the results in 61 cases Ann Surg, 135: 433, 1951.
80 LINTON, ROBERT R, HARDY, IRAD B, JR AND VOLWEILER, WADE. Portacaval shunts in the treatment of portal hypertension Surg, Gyn & Ob, 87: 129, 1948
81 MAHONEY, EARLE B. AND HOGG, LEWIS Congenital stricture of the portal vein Arch Surg, 61: 713, 1950
82 MALLORY, TRACY B: Case 20521 Mass Gen Hospital, New Eng J Med, 211: 1215, 1934
83 MANN, FREDERICK C. Studies in the physiology of the liver I Technic and general effects of removal Am J Med Sc, 161: 37, 1921
84 MARKOWITZ, J · Experimental Surgery, 2nd ed Baltimore, The Williams & Wilkins Co, 1949
85 MARKOWITZ, J, RAPPAPORT, A M AND SCOTT, A C Prevention of liver necrosis following ligation of the hepatic artery Proc Soc Exper Biol & Med, 70: 305, 1949
86 MARKOWITZ, J AND RAPPAPORT, A M · The hepatic artery Physiol Rev, 31: 188, 1951
87 DE MARTEL, M F. Brief summary of technic of Eck operation Rev de chir, 42: 1181, 1910
88 MAYO, WILLIAM J The surgical treatment of hepatic cirrhosis Ann Surg, 80: 419, 1924
89 McINDOE, ARCHIBALD H Vascular lesions of portal cirrhosis Arch Path, 5: 23, 1928
90 McLEOD, J J R AND PEARCE, R. G The outflow of blood from the liver as affected by variations in the condition of the portal vein and hepatic artery Am J Physiol, 35: 87, 1914
91 McMICHAEL, JOHN The portal circulation J Physiol, 75: 241, 1932
92 McMICHAEL, JOHN The pathology of hepatolienal fibrosis J Path & Bact, 39: 481, 1934
93 MEURSING, FOKKE Over een zeldsame anastomosose tusschen de vena porta en de vena cava Nederl tijdschr v geneesk, 2: 1678, 1912

50 FLEROW. Quoted by J. L. SPIWAK: The surgical technique of abdominal operations. Chicago, S. B. de Bour, 1941

51. FRERICHS, FRIEDRICH THEODOR A clinical treatise on diseases of the liver, 2 vols. London, New Sydenham Society, 1860–61.

52. GARDNER, RICHARD F., LEEDS, FRANK H. AND FREEMAN, NORMAN E. Experimental portacaval anastomosis. Calif. Med., 69: 1, 1948

53 GARLOCK, JOHN H AND SOM, MAX L : Further observations on packing of the mediastinum for esophageal varices J. Thoracic. Surg , 19: 572, 1950.

54 GERBODE, FRANK AND HOLMAN, EMILE: Experiences with the operative correction of portal hypertension Am J Surg , 82: 58, 1951.

55. GILBERT, A. AND WEIL, ÉMILE: Sur la tension des liquides ascitiques Soc de Biol , p 511, June 17, 1899

56. GILFILLAN, RUTHERFORD S . Anatomic study of the portal vein and its main branches Arch Surg , 61: 449, 1950.

57. GRANT, JOSEPH L , FITTS, WILLIAM T. AND RAVDIN, ISIDOR S Aneurism of the hepatic artery. Report of two cases and a consideration of surgical treatment. Surg , Gyn. & Ob , 91: 527, 1950

58 GRAY, HOWARD K. AND WHITESELL, FRANK B Hemorrhage from esophageal varices. Surgical management Ann Surg , 132: 798, 1950

59 GRINNELL, ROBERT S Omentopexy in portal cirrhosis of the liver with ascites. A review of twenty-three cases Ann Surg , 101: 891, 1935

60 GUNN, K E L G Portal and ovarian vein anastomosis Roy. Acad of Ireland, 8: 12, 1911.

61. HABERER, HANS *Experimentelle Unterbindung der Leberarterie Arch f klin* Chir , 78: 557, 1905

62 HARPER, HAROLD A , GARDNER RICHARD E , JOHANSEN, ROBERT, GALANTE, MAURICE AND MCCORKLE, H J. Amino acid tolerance in experimental portacaval anastomosis Surgery, 29: 210, 1951

63 HART, CARL Ueber die Bedeutung der Pfortadersklerose Arch f klin Chir , 118: 337, 1921

64. HERRICK, FREDERICK C An experimental study into the cause of increased portal pressure in portal cirrhosis J Exper Med , 9: 93, 1907

65. HIGGINS, GEORGE M AND MURPHY, GEORGE T I. Experimentally induced localized inflammatory reactions in the liver Arch Path , 9: 659, 1930

66. JULIAN, ORMAND AND FILDES, CHARLES E · Shunt operations for esophageal varices Surg Clin N Am , February, 229, 1951

67. KELTY, R H , BAGGENSTOSS, A H AND BUTT, H R The relation of the regenerated liver nodule to the vascular bed in cirrhosis Gast , 15: 285–295, 1950

68. KERSCHNER, DAVID, HOOTON, T. CAMPBELL AND SHEARER, E M Production of experimental portal hypertension in the dog Anatomy in the hepatic veins in the dog Arch Surg , 53: 425, 1946

vein Its relation

LOUISE Selective
enhagen, 1948
l'étude du syn-
'ascite, Paris, G

Steinheil, 1913

116 RIENHOFF, W. F, JR. Reported, Meet Soc. Univ. Surgeons, Baltimore, February 8, 1952.
117. ROLLESTON, H. D. AND FENTON, W. J., On the cirrhotic liver. Birmingh Med. Reb, 40: 193, 1896
118. ROMMELAERE, M: La pathologie de la veine porte. Bull. Acad roy de méd. de Belg, 17: 587, 1903
119 ROSENSTEIN, P Ueber die Behandlung der Leberzirrhose durch Anlegung einer Eck'schen Fistel. Arch f. klin. Chir, 98: 1082, 1912
120 ROUS, PEYTON AND LARIMORE, LOUISE D.: Relation of the portal blood to liver maintenance. A demonstration of liver atrophy conditional on compensation. J Exper. Med, 31: 609, 1920
121 ROUSSELOT, LOUIS M.: The role of congestion (portal hypertension) in so-called Banti's syndrome A clinical and pathologic study of thirty-one cases with the late results following splenectomy. J. A. M. A, 107: 1788, 1936
122. ROUSSELOT, LOUIS M. The late phase of congestive splenomegaly (Banti's syndrome) with hematemesis but without cirrhosis of the liver. Surgery, 8: 34, 1940
123. ROUSSELOT, LOUIS M.: Combined (one stage) splenectomy and portacaval shunts in portal hypertension With observations on venous shunts in the post splenectomy patient with recurring hemorrhage J. A M A, 140: 282, 1949
124. ROUSSELOT, LOUIS M. AND THOMPSON, WILLIAM P.: Experimental production of congestive splenomegaly Proc Soc. Exper. Biol. & Med., 40: 705, 1939
125 ROUSSELOT, LOUIS M.: Autogenous vein grafts in splenorenal anastomosis Surgery (in press)
126 SCHAFER, PAUL W AND KITTLE, C FREDERICK Partial esophagogastrectomy in the treatment of esophagogastric varices Arch Surg, 61: 235, 1950
127. SCHIFF, L Symposium on analysis and interpretation of symptoms; hematemesis and melena Clinics, 2: 1542, 1944.
128 SCHIFF, M: Ueber das Verhaeltnis der Lebercirculation zur Gallenbildung. Zentralbl med Wissensch, 8: 115, 1863
129 SCHMID, JULIUS Beeinflussung von Druck und Stromvolumen in der Pfortader durch die Atmung and durch experimentelle Eingriffe Pfluegers Arch. f Physiol, 126: 165, 1909
130 SENGSTAKEN, ROBERT W AND BLAKEMORE, ARTHUR H. Esophageal balloon tamponage for the control of hemorrhage for esophageal varices Ann Surg, 131: 781, 1950
131 SHERLOCK, S AND WALSHE, V. The use of a portal anastomotic vein for absorption studies in man Clin. Sc, 6: 113–121, 1948.
132 SIMONDS, J P AND BRANDES, W W The effect of obstruction of the hepatic veins on the systemic circulation Am J Physiol, 72: 320, 1925
133 SMITH, RICHARD M AND FARBER, SIDNEY Splenomegaly in children with early hematemesis J Pediat., 7: 585, 1935
134 SNELL, ALBERT M Clinical aspects of portal cirrhosis Ann Int Med, 5: 338, 1931
135 SOLOWIEFF, ALEXANDER: Veraenderungen in der Leber unter dem Einflusse kuenstlicher Verstopfung der Pfortader. Virch Arch f path Anat, 62: 195, 1875
136 SOM, MAX L AND GARLOCK, JOHN H New approach to the treatment of esophageal varices J A M A, 135: 628, 1947

94. MILNES, ROGER F AND CHILD, CHARLES G , III: Acute occlusion by ligature of the portal vein in the *Macacus rhesus* monkey. Proc. Soc. Exper. Biol. & Med , 70: 332, 1949.

95 MINO, ROBERT A , MURPHY, ARTHUR I. AND LIVINGSTON, ROBERT G : Sarcoidosis producing portal hypertension Treatment by splenectomy and splenorenal shunt. Ann Surg., 130: 951, 1949

96. MOERSCH, HERMAN J.. The treatment of esophageal varices by injection of a sclerosing solution. J. Thorac Surg , 10: 300, 1940

97. MORISON, RUTHERFORD · A case of ascites due to liver cirrhosis treated by operation Ann. Surg., 38: 361, 1903.

98. MOSCHOWITZ, E : Pathogenesis of splenomegaly in hypertension of portal circulation; "congestive splenomegaly" Med , 27: 187-221, 1948.

99 NEWMAN, H. F AND COHEN, I B Estimation of the portal circulation time in man J. Lab and Clin Med , 34: 674, 1949

100 ORE, M · Comptes Rend. Acad Sc , 42: 487, 1856.

101. OSLER, WILLIAM On splenic anaemia Am. J Med. Sc , 119: 54, 1900

102 PATEK, ARTHUR J , JR , POST, JOSEPH, RATNOFF, OSCAR D , MANKIN, HAROLD AND HILLMAN, ROBERT W. Dietary treatment of cirrhosis of the liver J. A. M. A , 138: 543, 1948

103. PEAN: Gaz. d'Hop , No 84, 1876

104. PEMBERTON, JOHN DEJ Results of splenectomy in splenic anemia, hemolytic jaundice and hemorrhagic purpura Collect Papers, Mayo Clin , 23: 639, 1931

105. PHEMISTER, DALLAS B AND HUMPHREYS, ELEANOR M Gastro-esophageal resection and total gastrectomy in the treatment of bleeding varicose veins in Banti's syndrome Ann Surg , 126: 397, 1947

106. PINCHANCOURT, MARCEL Contribution à l'étude du syndrome d'hypertension portale De la tension des liquides d'ascite Paris, G Steinheil, 1913

107. PICK, L Ueber totale hämangiomatose Obliteration des Pfortaderstammes und ueber hepatopetale Kollateralbahnen Virch Arch f path Anat , 197: 490, 1909.

108 POWER, WILLIAM Contributions to pathology, case IV—enteritis—varicose veins of the aesophagus Maryland Med & Surg J , 1: 316, 1840

109. PREBLE, ROBERT B Conclusions based on sixty cases of fatal gastro-intestinal hemorrhage due to cirrhosis of the liver Am J Med Sc , 119: 263, 1900

110. PRESHAW, D E , LARGE, ALFRED AND JOHNSON, ARTHUR F Effect of portacaval venous shunts on sulfobromophthalein (bromsulphalein) retention Arch Surg , 62: 801, 1951

111 RAIKEM, A F J Mem de l'Acad roy de méd de Belg , 1: 38, 1848

112 RATNOFF, O. D AND PATEK, ARTHUR J , JR Natural history of Laennec's *cirrhosis* Medicine, 21: 207, 1942

113 REYNOLDS, JOHN T AND SOUTHWICK, HARRY W Portal hypertension Use of venous grafts when side to side anastomosis is impossible Arch Surg , 62: 789, 1951

114 RICKETTS, WILLIAM E. AND KIRSNER, JOSEPH B . "Latent" portal cirrhosis Gastroent , 17: 184, 1951

115 RI

Pheochromocytoma

HENRY ARANOW, JR., M.D., MED SC.D.

Pheochromocytomas are tumors of chromaffin tissue which most frequently arise in and around the adrenal glands, but may be found in any of the widespread locations from neck to pelvis in which chromaffin tissue is present during early life. In recent years, interest in these comparatively rare neoplasms has been heightened by the development of new techniques for their identification, and better regimens for their removal. When unrecognized, the potent secretions elaborated by these tumors usually kill the individual harboring them.

Arterial hypertension is almost invariable at some time during the disease in patients with pheochromocytomas. Blood pressure elevation may be intermittent and paroxysmal, but usually it is continuous. Many of the pharmacological and physiological actions of the pressor principles of these new growths have been analyzed. It is probable that studies of patients with chromaffin tumors may lead to better understanding of the mechanisms in hypertensive vascular disease. This probability is increased by the repeated observation that the syndrome in some patients with pheochromocytomas is frequently indistinguishable from that seen in hypertension.

In this paper, primary emphasis will be given to physiological and pharmacological studies. A number of reviews with other orientations have been published, and Graham's excellent bibliography includes two hundred and forty-nine references.

INCIDENCE

The incidence of pheochromocytoma cannot be stated with certainty. Graham estimated that between six and eight hundred patients in the United States die annually from this disease, on the basis of his finding "pheochromocytoma was encountered eight times (0 47 per cent) in 1,700 unselected patients with hypertension subjected to bilateral lumbodorsal splanchnicectomy" and the reported 175,000 annual deaths from hypertension The statistical weakness of such a calculation needs no comment, but it is indubitable that the three hundred odd reported chromaffin tumors represent but a small fraction of the actual number. Every author who has written any sort of review of the subject has been able to add several previously unreported cases and four such are included below.

Unless a pathologist collects a considerable number, or studies an extraordinary case, the entity is now known well enough that he has little in-

137. STAHL, GEORG ERNEST De vena portæ, porta malorum hypochondriaco-splenetico-suffocativo-hysterico-colico-haemorrhoidariorum. [Recusa] Halœ ad Salam, sumpt. J. C. Hendelii, 1751.
138. TALMA, S · Chirurgische Oeffnung neuer Seitenbahnen fuer das Blut der Vena Porta Berl klin. Wchnschr, 35: 833, 1898
139 THOMPSON, WILLIAM P. The pathogenesis of Banti's disease. Ann Int. Med., 14: 255, 1940
140. THOMPSON, WILLIAM P., CAUGHEY, JOHN L, WHIPPLE, ALLEN O AND ROUSSELOT, L M Splenic vein pressure in congestive splenomegaly (Banti's syndrome) J Clin Invest, 16: 571, 1937.
141. VIDAL, E · Discussion of Eck operation Rev. de Chir, 42: 1181, 1910
142 VILLARET, MAURICE · *Contribution à l'étude du syndrome d'hypertension* portale Paris, G. Steinheil, 1906
143. WAKIM, KHALIL G. AND MANN, FRANK C. Effect of experimental cirrhosis on the intrahepatic circulation of blood in the intact animal Arch. Path, 33: 198, 1942
144 WANGENSTEEN, OWEN W Discussion of D B. PHEMISTER AND E. M. HUMPHREYS: *Gastro-esophageal resection and total gastrectomy in the treatment* of bleeding varicose veins in Banti's syndrome. Ann Surg, 126: 397, 1947.
145 WARTHIN, ALDRED SCOTT The relation of thrombophlebitis of the portal and splenic veins to splenic anaemia and Banti's disease Inter Clin, 20th series, 4: 189, 1910
146. WELLS, SPENCER Remarks on splenectomy with a report of a successful case. Brit Med J., II: 796, 1888
147 WELLS, SPENCER Med Times & Gaz, 1: 2, 1866
148 WELLS, SPENCER Diagnosis and surgical treatment of abdominal tumours Phila, P Blakiston Sons, 1885
149 WHIPPLE, ALLEN O. The medical-surgical splenopathies Introduction Bull N Y Acad Med, 15: 174, 1939
150 WHIPPLE, ALLEN O The problem of portal hypertension in relation to the hepatosplenopathies Ann Surg, 122: 449, 1945
151 WHITE, FRANKLIN W AND CHALMERS, THOMAS C The problem of gross hematemesis in general hospital A study of 400 consecutive cases Tr Ass Am. Phys, 61: 253, 1948
152 WOMACK, NATHAN A The surgery of portal hypertension P 692 Operative Technic Ed Warren Cole, Appleton Century-Crofts, Inc N Y, 1949
153. WOMACK, NATHAN A Discussion of H K GRAY AND F B WHITESELL, JR Hemorrhage from esophageal varices Surgical management Ann Surg, 132: 808, 1950
154 ZAMCHECK, NORMAN, CHALMERS, THOMAS C, WHITE, FRANKLIN W AND DAVIDSON, CHARLES S. The bromsulphalein test in the early diagnosis of liver disease in gross upper gastro-intestinal hemorrhage Gastroent, 14: 343, 1950

right heart catheterization and application of the Fick principle; arterial pressures were recorded directly with a Hamilton manometer. Their data conclusively confirmed the fact that physiologic doses of epinephrine had an overall *vasodilator* action, and *lowered* the total peripheral resistance. An increase in mean arterial pressure was constantly recorded, but this was due to an augmentation in cardiac output which more than overbalanced the peripheral vasodilation. These observations were repeated by DeLargy and his collaborators. There is evidence, obtained by intravenous infusion of epinephrine in a patient whose blood pressure was constantly elevated by a pheochromocytoma which contained predominantly epinephrine, that doses of epinephrine well beyond the physiologic range may have vasoconstrictor action.

During the infusions of epinephrine an increase in basal oxygen consumption was constantly observed, which accorded with earlier findings by many workers

Amounts of epinephrine only about half as great as those required to raise the mean blood pressure were shown by Cori and Buchwald to raise the level of circulating blood glucose. The increase in the amount of glucose delivered to the body by the liver in response to epinephrine infusions has been more quantitatively examined by Bearn, Billing, and Sherlock who also noted that the hormone increased hepatic blood flow significantly

Cardiac output is increased by epinephrine because of a more complete systolic ejection and an acceleration of the heart. Epinephrine increases coronary blood flow in normal hearts, but this augmented flow falls considerably short of the greater needs of the myocardium Indeed, Raab has stated, "Epinephrine is capable of causing myocardial anoxia as a specific metabolic effect regardless of hemodynamic conditions and regardless of the volume of coronary flow"

Epinephrine diminishes renal blood flow without causing much change in glomerular filtration rate, so that the filtration fraction rises. In addition, Nickel and his colleagues have found that in man there is an increased tubular reabsorption of sodium and potassium, and a decreased reabsorption of water This can be observed after doses of epinephrine too small to affect the pulse rate or blood pressure; larger doses have similar effects.

Vogt's observation that epinephrine plays a major role in the chain of events which leads to an increased output of adrenal cortical steroids has been amplified by a number of workers.

Finally, Wada has recently elucidated the sudorific action of epinephrine on human sweat glands, and has furnished a key to the sweating seen in many patients with pheochromocytomas. The well-established cholinergic nature of the innervation of the sweat gland had led to some rather involved hypotheses to account for what appeared to be an anomaly.

centive to publish. Although clinicians are more likely to report single cases diagnosed ante-mortem, in more than half of the instances, first recognition occurs in the autopsy room.

Pheochromocytomas have been found with approximately equal frequency in either sex, and they occur in patients ranging in age from five months to 72 years. More than 70 per cent have been identified between the ages of 20 and 60.

Physiology

Like all neoplasms, pheochromocytomas may cause symptoms by displacement or compression of adjacent organs, but the characteristic dramatic disturbances are caused by their endocrine effects

Until 1949 it was assumed that the hormone in pheochromocytomas was epinephrine Extracts of many tumors had the biological and chemical characteristics of this catechol. In that year Holton, using more discriminating biological techniques, and Goldenberg et al., employing a new *chromatographic chemical method, demonstrated that pheochromocytomas* contain significant amounts of norepinephrine as well as epinephrine. In the large number of tumors which have now been analyzed by Goldenberg and his co-workers, norepinephrine has been shown to constitute from 14 to 97 per cent of the total pressor catechol content The percentage of norepinephrine has been as great, and usually greater, than they found in the normal adrenal medulla

The pressor activities of the tumor extracts, as well as their other effects are completely accounted for by their epinephrine and norepinephrine content. One may properly infer, therefore, that the syndrome in patients with pheochromocytomas represents the effect of the intermittent or more or less continuous infusion of varying mixtures of these catechols.

Epinephrine

The pharmacological properties of epinephrine have been extensively studied. Certain aspects of its hemodynamic actions, however, have been insufficiently emphasized. There is general awareness of the fact that more than minute doses of epinephrine tend to raise the mean blood pressure of man, this effect is observed with subcutaneous, intra-muscular, and intravenous routes of administration In spite of studies by Cannon and Lyman, by Starr et al., and by Ranges and Bradley, it has often been incorrectly deduced that this pressor effect was due to an overall vasoconstrictor action of epinephrine.

The hemodynamic effects *of continuous intravenous infusions of physiologic* doses of epinephrine and norepinephrine have been recently studied in man by Goldenberg, Pines, et al. Cardiac output was determined by

right heart catheterization and application of the Fick principle; arterial pressures were recorded directly with a Hamilton manometer. Their data conclusively confirmed the fact that physiologic doses of epinephrine had an overall *vasodilator* action, and *lowered* the total peripheral resistance. An increase in mean arterial pressure was constantly recorded, but this was due to an augmentation in cardiac output which more than overbalanced the peripheral vasodilation. These observations were repeated by DeLargy and his collaborators. There is evidence, obtained by intravenous infusion of epinephrine in a patient whose blood pressure was constantly elevated by a pheochromocytoma which contained predominantly epinephrine, that doses of epinephrine well beyond the physiologic range may have vasoconstrictor action.

During the infusions of epinephrine an increase in basal oxygen consumption was constantly observed, which accorded with earlier findings by many workers

Amounts of epinephrine only about half as great as those required to raise the mean blood pressure were shown by Cori and Buchwald to raise the level of circulating blood glucose The increase in the amount of glucose delivered to the body by the liver in response to epinephrine infusions has been more quantitatively examined by Bearn, Billing, and Sherlock who also noted that the hormone increased hepatic blood flow significantly.

Cardiac output is increased by epinephrine because of a more complete systolic ejection and an acceleration of the heart. Epinephrine increases coronary blood flow in normal hearts, but this augmented flow falls considerably short of the greater needs of the myocardium Indeed, Raab has stated, "Epinephrine is capable of causing myocardial anoxia as a specific metabolic effect regardless of hemodynamic conditions and regardless of the volume of coronary flow."

Epinephrine diminishes renal blood flow without causing much change in glomerular filtration rate, so that the filtration fraction rises. In addition, Nickel and his colleagues have found that in man there is an increased tubular reabsorption of sodium and potassium, and a decreased reabsorption of water. This can be observed after doses of epinephrine too small to affect the pulse rate or blood pressure, larger doses have similar effects

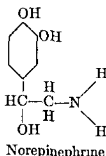
Vogt's observation that epinephrine plays a major role in the chain of events which leads to an increased output of adrenal cortical steroids has been amplified by a number of workers.

Finally, Wada has recently elucidated the sudorific action of epinephrine on human sweat glands, and has furnished a key to the sweating seen in many patients with pheochromocytomas The well-established cholinergic nature of the innervation of the sweat gland had led to some rather involved hypotheses to account for what appeared to be an anomaly.

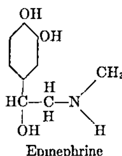
In the conscious normal human subject, an infusion of epinephrine at a rate sufficient to produce a moderate rise in mean arterial pressure produces palpitation, tremors, anxiety, sweating, hyperpnea, and other disturbing symptoms

Norepinephrine

Norepinephrine is a primary amine, and it differs from epinephrine by having two hydrogen atoms on the nitrogen atom, rather than one hydrogen atom and a methyl group.



Norepinephrine



Epinephrine

Luduena et al. found that almost all of the pharmacological activity of the racemic form of norepinephrine resided in the levo-rotatory isomer, and Lands and his colleagues have studied the comparative pharmacology of the series of compounds obtained by substituting various alkyl groups for one of the amino hydrogens

The finding that norepinephrine is released when adrenergic nerves are stimulated, and its presence in the normal adrenal medulla indicate that the physiological importance of the primary amine is probably as great as that of its methylated congener

Goldenberg et al. demonstrated that, in sharp contrast to epinephrine, intravenously infused norepinephrine raises the arterial blood pressure by causing peripheral vasoconstriction with little effect on cardiac output A sharp rise in total peripheral resistance is its most striking effect, mainly caused by narrowing of the enormous vascular bed of the skeletal muscles In the otherwise untreated subject, the blood pressure elevation is usually accompanied by a bradycardia of reflex origin, the latter can be abolished by atropine and an even more striking pressor effect observed.

Lu and Melville observed that norepinephrine had a marked direct stimulatory action on the myocardium, it is probable that this leads to an even greater relative hypoxia of the heart muscle fibers than is caused by epinephrine.

Norepinephrine has a much smaller effect than epinephrine on hepatic blood flow and glycogenolysis On the basis of gravimetric dosage, nor-

epinephrine is only one-fourth to one-eighth as active in raising blood sugar as is epinephrine.

The action of norepinephrine on the kidney is qualitatively and quantitatively similar to that of epinephrine; the same increase in filtration fraction and of tubular sodium and potassium reabsorption, and the same diminution in total renal blood flow and tubular water reabsorption was found.

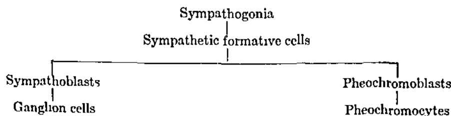
Norepinephrine stimulates only slightly the production of adrenocorticotrophic hormone by the anterior hypophysis. Its effect on human sweat glands is unknown.

When norepinephrine is infused intravenously into the conscious human subject, the absence of symptoms is strikingly at variance with the syndrome which follows epinephrine administration. Major rises in mean arterial pressure are often accompanied by no more than slight awareness of a slow, forceful beating of the heart.

Such differences in the actions might lead one to wonder whether the finding of Goldenberg et al that "U.S P. Epinephrine" contained, on the average, about 18% norepinephrine will require the repetition of the many studies on the action of epinephrine, since most experimenters used "epinephrine" of animal origin, rather than synthetic l-epinephrine. Fortunately it appears that this will not be necessary. It has been shown by Goldenberg and his colleagues, and by DeLargy's group, that norepinephrine must constitute considerably more than half of the mixture before significant qualitative changes from the established actions of "epinephrine" become manifest.

Pathology

The tumors are made up of mature chromocytes They were named pheochromocytomas because they stain brown in chromic salt solutions. No recent evidence has necessitated changes in the schema presented by Rabin which is given below. The precursors of both the neural and endocrine elements are derived from the neural crest, from which they migrate and split off.



Tissue of this origin is found as high as the neck and as low as the uterine

cervix, but about 85 per cent of tumors which have produced symptoms during life have been found in the adrenal areas. Hofbauer has recently suggested that the chromaffin elements in the upper vagina and cervix may play an endocrine role in some of the vascular disturbances in pregnancy.

Pharmacologically active tumors have been identified in the organ of Zuckerkandl at the bifurcation of the aorta, in the celiac ganglion, and in the thorax. In almost 20 per cent of cases the tumors are multiple, and in about half of these are bilateral The first reported case of pheochromocytoma occurred in an 18-year-old girl who was shown by Frankel to have bilateral new growths. It is my experience that young people are likely to have multiple tumors.

Microscopically the tumors are usually composed of polymorphic and polyhedral cells The cells may be arranged in alveolar masses, or in long intertwining columns, at times they show pseudo-rosette formation around vascular spaces. The columns or masses are separated by fibrous tissue septa which are often so vascular as to resemble sinusoids. Extreme vascularity is characteristic, and most large growths show areas of hemorrhagic necrosis as well as cystic changes in areas of old hemorrhage. A patient admitted to the Presbyterian Hospital with abdominal pain and shock succumbed to a massive retroperitoneal hemorrhage which was shown at postmortem examination to have originated in an unsuspected pheochromocytoma, McFarland and Bliss have published the successful management of a similar case

In most instances the cells contain abundant, finely granular cytoplasm, and the nuclei tend to be round or oval with a readily identifiable nucleolus. Multinucleated giant cells, and cells arranged in syncytial sheets are seen occasionally Mitoses are of moderate frequency Most pathologists agree that the diagnosis of malignancy cannot be made with certainty from the histological characteristics of the tumor, but depends on metastases or invasion of adjacent structures

At times cortical or neural elements may be present in small numbers, and the pathological report of our most recently excised pheochromocytoma reads: "... *Nests of cells resembling cortical cells are found within the tumor itself* An interesting feature is the presence, in one area of the tumor, of ganglion cells and clusters of small, lymphocyte like cells which are probably *sympathicoblasts* ."

The nature of the neuronal connections of pheochromocytomas has not been studied quantitatively or precisely, so that we have little knowledge of the role of neural mechanisms in the regulation of the release of their secretions.

As would be anticipated from their high degree of vascularity, the tumors

often derive large arteries from the adjacent aorta, adrenal, or renal artery and the prominent veins may empty directly into the vena cava, or the renal and adrenal veins.

Lymphatic connections are inconspicuous; in instances of malignancy, spread is hematogenous or by direct invasion

The tumors may range in size from nodules less than one centimeter in diameter to masses more than 10 centimeters in diameter.

Some pheochromocytomas which have been asymptomatic are found at postmortem examination. Apparently they play no role in causing death, and may do no harm nor cause any lesion.

A number of patients die suddenly during anesthesia or minor operative procedures as a result of sudden discharges from unsuspected pheochromocytomas. Findings at postmorten examination in such cases are highly variable, although many show pulmonary edema and varying degrees of congestion of the viscera. Similar changes may be encountered in patients who succumb during the operative removal of a chromaffin tumor which has caused paroxysmal hypertension If, as is frequently the case, death is preceded by a period of shock, the nonspecific pathologic changes of shock are often recorded

Finally, in patients with persistent elevation of arterial pressure, any or all of the pathologic findings of essential and malignant hypertensive vascular disease may be present.

The thickening and hyalinization of the renal arterioles, so constant in patients dying of hypertensive disease, was observed in Frankel's original patient, and in many cases of pheochromocytoma since In Green's review of patients with pheochromocytoma and persistent hypertension, seven of the 16 patients whose kidneys were studied showed such changes In the patient reported by Washington et al, a necrotizing arteriolitis identical with that characteristic of malignant hypertension was found not only in the kidneys but in the pancreas

Hemorrhage and thrombosis in damaged cerebral vessels are frequent terminal events in the course of patients with pheochromocytomas More than a fourth of the group of patients reported by Graham who died while harboring untreated tumors succumbed to cerebral vascular accidents.

Retinal vascular damage is often severe, and it is my impression that it is of greater degree than that usually seen in patients with blood pressure elevations of comparable magnitude and duration caused by essential hypertension

Cardiomegaly is almost the rule in patients who die with persistent hypertension caused by pheochromocytoma, and it is often associated with varying degrees of myocardial fibrosis Congestive failure ranks with cerebral vascular accidents as a cause of death in patients with untreated

tumors. The myocardial hypertrophy is often greater than would have been anticipated from the known duration of the hypertension and the level of the blood pressure. This excessive hypertrophy accords well with the effects of norepinephrine and epinephrine on the myocardium. The patient reported by Washington et al. had hypertension for only seven months, but his heart weighed 630 grams

Atherosclerosis of the coronary arteries, as well as generalized arteriosclerosis are frequent, but comparable to that found in hypertensive patients of similar age.

Clinical Picture

The best known manifestation of pheochromocytoma is the "adrenal sympathetic syndrome." In its classical form it is caused by the sudden outpouring of a mixture of epinephrine and norepinephrine, in which the effect of the former is dominant. The constant feature of this syndrome is a sudden increase in blood pressure, this is usually accompanied by other evidence of sympathetic activity such as palpitation, tachycardia, sweating, generalized trembling, blanching (often followed by flushing) of the skin, coldness of the hands and feet, hyperglycemia, and glycosuria. In addition, there is often pounding headache, or abdominal and precordial pain. Any combination of these may characterize the attacks of a given patient The original papers of Labbe et al., and of Vaquez and Donzelot contain descriptions which are most vivid, a translation from the latter is given below:

"The patient was a 37 year old man whose general health was excellent prior to the paroxysmal attacks which had begun six months before

"Suddenly, during either exertion or emotion, or at times without the least ap- [illegible]

[illegible]

short interval, by neck and chest pain of anginal character, and, finally, by a frightful headache During the rapid development of this painful syndrome no nausea, vomiting, convulsive phenomena, nor impairment of consciousness appeared The crisis lasted several minutes, and the pains in the extremities, abdomen, chest, and head were so severe, as to be intolerable, and the patient lived in continual fear of their return.

"These attacks reoccurred at times in their complete form, at others in an attenuated form, in the three preceding weeks their frequency had increased so that several were being experienced in each 24 hours

"By good fortune we were present, in the course of our examination, during the [illegible] We had already examined that patient's heart, which [illegible] pressure to be 140/80 [illegible] entire skin blanched, [illegible] celerated so abruptly [illegible] ompletely changed in a few seconds; the rate had increased from [illegible] to [illegible] per minute, and in place of the

formerly normal second sound one heard a veritable aortic clangor. The sphygmomanometer was still on his arm, and we quickly took the pressure which had risen from 140/80 to 300/170!

"The hypertensive crisis reached its peak during the third minute, and then began to subside, we recorded the successive blood pressure values of 210/130, 180/110, and, in the seventh minute, 140/90."

Such symptoms and signs are those which would be expected from generalized discharge of the sympatho-adrenal system. Since such discharges can be initiated centrally in the absence of a pheochromocytoma, it is not surprising that identical changes can be caused by overwhelming anxiety, or by a neoplasm in the hypothalamic region. In a case described by Penfield, the syndrome was produced by a ball-valve tumor of the third ventricle.

The manifestations of anxiety attacks have been so suggestive of pheochromocytoma that a number of patients have been operatively explored even when *all* of the pharmacological tests have been negative. I know of no instance of this sort in which a chromaffin neoplasm has been found. On the other hand, even when psycho-emotional factors of great importance are self-evident, it is not safe to assume that they alone are responsible for the outbursts

J B C (P H #038950) was a 23-year-old single white dockhand who was referred to the Medical Clinic for study because of increasingly severe "attacks," weakness, palpitation, occipital headaches, nausea and vomiting He was mentally retarded and had many phobias and evidences of insecurity About a year earlier he began to have almost daily episodes of vague weakness precipitated by exertion and lasting only a few seconds The bouts increased in frequency and severity Seven months prior to coming to the clinic they were accompanied by occipital headaches which lasted two to three hours, and by palpitation and sweating Later he had several each day One month before admission the attacks included nausea and vomiting which were not prevented by antacid therapy In the few months preceding his first visit the attacks began without evident precipitating cause

Physical examination on admission revealed the patient to be very tense The skin was moist and the blood pressure was 138/94 There were no other noteworthy findings

Laboratory studies revealed normal hemogram, normal x-rays of heart, lungs, and gastrointestinal tract, electroencephalogram and radioactive iodine tracer uptake were within normal limits, urinalysis was normal save for one plus albumin

The diagnosis of pheochromocytoma was never seriously entertained, and no attempt to exclude it was made

During his workup the patient revealed that he had such a phobia about signing his name that marriage was unthinkable because he could not bring himself to sign the license application He was seen by a psychiatrist who stated that the "spells" represented anxiety attacks of an acute nature occurring in an individual with inadequate personality and mental deficiency

During one of his attacks he was admitted to the Overnight Ward Blood pressure on admission was 150/100 Sedatives were administered and, after consultation, it

was agreed that he should be referred to the Mental Hygiene Clinic of his local city hospital for therapy.

Less than two weeks later he had a convulsion at home and died Autopsy by the Medical Examiner disclosed a pheochromocytoma of the right adrenal gland which weighed 150 grams Goldenberg's analysis showed 1.17 milligrams of norepinephrine and 2 09 milligrams of epinephrine per gram of tumor.

In many patients, attacks bear a fairly constant relationship to some provocative factor. These factors vary widely from patient to patient and include postural change, emotional upsets, physical exertion, hyperventilations, exposure to cold, and abdominal massage. In many, however, the paroxysms are quite unpredictable and can be regularly reproduced only by the exhibition of some of the pharmacological agents discussed under "Diagnosis"

The tumors of patients who had paroxysmal rather than maintained hypertension have contained the same widely varying mixtures of epinephrine and norepinephrine. It is therefore not surprising that some patients with paroxysmal elevations of blood pressure show only parts of the multifacetted picture of the classical "adrenal-sympathetic syndrome".

DeVries and his colleagues described two patients with paroxysmal hypertension one of whom had persistent and the other paroxysmal diabetes, both were cured not only of paroxysmal hypertension but of diabetes when their pheochromocytomas were removed Calkins, Dana, and Howard mention a patient who, although he never showed an elevated blood pressure during six weeks of observation, was cured of diabetes by removal of a chromaffin tumor

Sudden blood pressure rises without accompanying hyperglycemia, and with only a few subjective disturbances, have been observed in other patients with pheochromocytomas

A rare but extremely interesting complication of the adrenal-sympathetic syndrome has been the appearance of shock after a prolonged paroxysm. This appears to be the clinical counterpart of the condition Freeman and his colleagues produced by the prolonged continuous infusion of epinephrine The woman studied by Engel, Mencher, and Engel had attacks which lasted as long as 36 hours. During such a period the arterial tension, which had been as high as 310/180, slowly declined to 76/60, and the temperature rose to 104°F Following successful operation all of her symptoms cleared The young soldier reported by Ferraro and Angle died in shock 14 hours after the start of an attack At the time of death his temperature had reached 106.4°F and the clinical picture suggested overwhelming infection. The only important postmortem finding was a 70 gram pheochromocytoma of the left adrenal gland

Fertig et al have indicated that a "paroxysmal nephritis" accompanying

a paroxysmal hypertension should point to the presence of a chromaffin tumor. The cylindruria, impaired excretion of phenolsulphonphthalein and albuminuria may change markedly toward normal between paroxysms; complete restoration often follows removal of the tumor

Although all of the clinical findings caused by pheochromocytoma can be accounted for by the known actions of epinephrine and norepinephrine, the factors which control their secretion are widely varying and poorly understood. Finding of a certain number of milligrams of each substance in a neoplasm removed at operation or found at postmortem examination, does not give the slightest inkling of the degree of physiologic disturbance produced by it, nor of the frequency or duration of episodes of "activity" of the tumor. Tumors of small size and modest pressor amine content may produce sudden death in one patient while another may have a growth containing more than 800 milligrams of epinephrine and carry it for years without symptoms

In a significant number of instances the pheochromocytoma is a purely incidental finding either at autopsy or at an operation performed for other reasons Re-examination of the history and careful scrutiny of the physical and laboratory findings after identification of the new growth may reveal no evidence of secretory activity of the pheochromocytoma Whether such activity might have been provoked by some of the pharmacological measures discussed below is unknown. Much could be added to our understanding of the problem by a meticulous comparison of the neural and vascular connections of dormant neoplasms and active and physiologically disturbing tumors

L W (P H #718506) was a 62-year-old Negro man with progressive pulmonary insufficiency caused by silicosis He was studied on the wards of the Presbyterian Hospital over a considerable period of time, blood pressure was never significantly above 120/80, and glycosuria was not observed In spite of the stresses imposed by increasing hypoxia and various studies of pulmonary function, no episodes suggesting excessive outpouring of epinephrine or norepinephrine were observed He eventually succumbed to his pulmonary disease, and at postmortem examination in the left adrenal we found a typical pheochromocytoma which measured 1 0 x 1 5 cm

In other patients, the induction of anesthesia or some minor trauma may cause the outpouring of a lethal quantity of the pressor amines, and later review of the available data often reveals nothing to suggest prior hypertension, either intermittent or continuous In the recorded anamnesis there is usually no mention of spells of palpitation, of sweating, or of headache This implies that the patient did not volunteer such data, since the examiner was not seeking it by specific questioning In Apgar and Papper's study of patients with pheochromocytomas who came to operation they

noted that more than 50 per cent of patients with previously undiagnosed pheochromocytomas died during or shortly after operation.

F. McI (P.H. #002011) was a 56-year-old white man admitted to the Neurological Institute with an eight months' history of gradually increasing weakness of the legs and numbness of the hands X-rays and myelographic studies led to the diagnosis of Klippel-Feil deformity of the cervical spine with probable herniation of several cervical intervertebral discs Medical history was otherwise unremarkable, and preoperative blood pressure was 148/95 Laminectomy was advised.

Thirty minutes after an otherwise uneventful induction of anesthesia with procaine, pentothal, nitrous oxide, and oxygen the patient's systolic blood pressure rose to 215 mm Hg and he developed what the anesthetist thought was an asthmatic attack Intravenous aminophyllin, Isuprel, and ephedrine were administered, but the blood pressure suddenly became unobtainable and death occurred before the laminectomy was started

Autopsy showed an enlarged heart which weighed 700 gm and contained an old, healed infarction There was a pheochromocytoma of the left adrenal gland which measured 2 0 cm x 3 5 cm The lungs showed pulmonary congestion, and the cervical cord compression was confirmed

It is of interest to contrast this patient with another followed at the Presbyterian Hospital for three and a half years.

K B (P H #910743) a separated, colored housewife was 35 years old in May 1948 when she came to the Admitting Ward because of sore throat and fever Physical examination revealed temperature 102 8°F , B P 120/80 There was acute follicular tonsillitis Abdominal palpation disclosed a firm, globular, non-tender mass the size of a tennis ball in the left upper quadrant The mass moved with respiration The uterus contained fibromyomata The patient was admitted to the Overnight Ward where a flat film of the abdomen revealed a rounded, homogeneous mass which obscured the left kidney shadow and pushed the stomach upward and the colon downward

The medical resident wrote, "For past two years or so she often awakens at night trembling all over; also complains of palpitation These complaints are usually relieved by vomiting She never has these attacks during the day Occasionally she complains of mild mid abdominal distress during one of these attacks Mass was subjected to massage with no change in blood pressure which remained constant in the 120/80 range Doubt hyperadrenalism " The consulting surgical resident added, "Agree that the best bet is pancreatic cyst However, absence of kidney shadow and firm nature of mass suggest solid tumor, possibly of kidney origin "

Before the studies suggested in the Medical Clinic were complete, the patient [illegible] to the Ear, Nose and Throat Service Admission blood pressure was [illegible] l anesthesia was uneventful [illegible] ed that the mass distorted the superior calyces of the left kidney, and lay medial to the splenic flexure of the colon The urologist suggested retrograde studies, but the surgical consultant concurred in the diagnosis of pancreatic cyst, and signed the patient in for laparotomy at the end of *June*, 1948

For various reasons the patient was unable to enter the hospital, and the retrograde studies were done in February, 1949 The urologist was still uncertain of the nature of the mass, and recommended peri-renal air insufflation

The patient did not return until September, 1951 At this time her complaint was right lower quadrant pain, and she was referred to the Gynecological Clinic, from which she entered the hospital for complete hysterectomy. Examination revealed the left upper quadrant mass as described in 1948, blood pressure of 150/100, and a fibroid uterus

The surgical resident was invited to examine the left upper quadrant mass at the time of the complete hysterectomy, which was described as uneventful He palpated the mass, and thought that it was a pancreatic cyst measuring about 8 5 cm. in diameter.

After a smooth convalescence, the patient was admitted a month later to the Surgical Service Blood pressure on admission was 124/86 and a glucose tolerance test was normal.

Induction of anesthesia for the laparotomy was unremarkable, and the mass was seen to arise from the inferior aspect of the left adrenal gland, and to receive a large arterial supply from the renal artery The surgeons were under the impression that they were dealing with an adrenal cortical neoplasm The operation proceeded without incident until the tumor was manipulated, when the blood pressure rose to 220/130. Following excision of the mass, the arterial tension fell to shock levels, but was promptly restored to normal with an infusion of norepinephrine

It was not until the tumor reached the Surgical Pathologist that the diagnosis of pheochromocytoma was made The total weight was 220 gm , and prompt analysis by Goldenberg revealed 4 06 milligrams of epinephrine per gram of tumor (86%) and 0 653 milligrams of norepinephrine per gram (14%).

Two months after operation the patient was completely well

Save for the initial notes, no further mention of attacks of nocturnal trembling and palpitation was made during her three and one-half years of intermittent observation. It was not until the diagnosis had been established that re-examination of the anesthesia record charted during hysterectomy revealed that the blood pressure had risen when the surgeon palpated the tumor In spite of the fact that she carried within her a veritable pharmacological bomb, no evidence of catechol discharge was observed until the mass was actually manipulated, only then did it give a hint of its physiologically malignant potentialities.

Since epinephrine and, to a much lesser extent, norepinephrine cause an increase in basal oxygen consumption, hypermetabolism frequently is a striking feature in patients with pheochromocytomas The increase in sensitivity to epinephrine of patients with thyrotoxicosis is well known, and the resemblance of the thyrotoxic state to one of increased sympathetic activity had been noted by many It is not surprising, therefore, that many patients with pheochromocytomas have been mistakenly thought to have hyperthyroidism. Thyroidectomy and antithyroid drugs have usually been of no benefit, and patients have died during thyroidectomies for the removal of normal thyroid glands McCullagh and Engel, Raab and Smithwick, and others have emphasized the metabolic features of pheochromocytomas Espersen and Dahl-Iversen gave methylthiouracil to a patient

with the adrenal-sympathetic syndrome who had previously shown temporary improvement after a partial thyroidectomy; they reported that the paroxysms were abolished by the drug during the month it was administered. The patient was then cured by operative removal of his tumor. This observation has not been confirmed, and may well have been fortuitous.

In a few instances, evidences of adrenal cortical hyperfunction have been prominent in patients with pheochromocytoma. In the child reported by Neff et al. these disappeared after the removal of a chromaffin tumor, and one must presume that the hyperadrenocorticism was secondary to overstimulation of the anterior pituitary by epinephrine.

Although many authors have thought that the paroxysmal attacks of the adrenal-sympathetic syndrome were the precursors of the persistent blood pressure elevation observed in the majority of patients with pheochromocytomas, a number of patients have been described whose hypertension remained purely paroxysmal for more than 40 years, and whose arterial pressures and blood sugar levels were quite normal during the intervals between attacks.

Until recently it was generally believed that the most frequent, as well as the most striking, syndrome presented by patients with pheochromocytomas was characterized by paroxysms of hypertension with normal blood pressure between attacks. Green reviewed 51 patients with adequate data on blood pressure and found that in 37 (more than 70 per cent) the blood pressure elevation was persistent rather than intermittent This is consonant with Smithwick's experience, as well as my own In most, but not all, of these patients careful questioning will elicit a history of symptomatic episodes during which a rise above the usual level of arterial pressure is suggested In the remainder, however, an exhaustive history and careful observation fail to disclose evidence of significant fluctuation of arterial tension

This latter group of patients so closely resemble those with essential hypertensive vascular disease that the correct diagnosis often is first made at *postmortem* examination or during routine exploration of the adrenal areas in the course of a lumbo-dorsal sympathectomy In only two of the 10 patients with chromaffin tumors in the adrenal area who were reported by Smithwick et al. was the diagnosis made prior to splanchnicectomy

When this syndrome occurs during pregnancy, the persistent hypertension is usually ascribed to a hypertensive toxemia More than 10 instances of pheochromocytoma associated with pregnancy have been published, and in several the pregnancy has been needlessly interrupted.

It seems probable that only the routine use of some pharmacological screening test will materially increase the correct preoperative diagnoses in patients with persistently elevated and relatively constant hypertension.

Patients with persistent hypertension caused by pheochromocytomas are subject to the same complications found in hypertensive disease of other etiology, but these tend to appear quite early in the course of the disease. Cerebral vascular accidents, retinopathy, myocardial infarction, cardiac failure, and uremia have been observed.

Diagnosis

The major diagnostic problems presented by chromaffin tumors fall into two divisions; First, it is necessary to distinguish paroxysmal increases in blood pressure, with or without associated findings, caused by hormonal discharge by pheochromocytomas from sudden hypertensive episodes of other cause. Second, a method to separate those patients with persistent hypertension secondary to pheochromocytoma from the great number of patients with essential and malignant hypertension must be found; in exceptional instances differentiation from Cushing's syndrome or chronic glomerulonephritis may be a problem.

The clinical, radiographic, and pharmacological findings of value in making these differentiations are described and evaluated in this section.

Whenever a patient gives a history of episodes of paroxysmal hypertension, the diagnosis of pheochromocytoma is usually entertained, but paroxysmal increases in arterial pressure are by no means pathognomonic *of pheochromocytoma. Some of the other conditions in which such par*oxysms have occurred are detailed in the monograph of Bernal and the more recent paper by Bauer and Belt. The activation of the sympathetic center in the hypothalamus either by strong psychoemotional stimuli or by adjacent neoplasms leads to a generalized sympatho-adrenal discharge which has most of the clinical and pharmacological characteristics of an attack produced by the outpouring of catechols from a chromaffin tumor. Less readily explicable paroxysms of hypertension may occur in eclampsia, in essential hypertension, in various central nervous system disorders such as epilepsy, tabes, cerebral vascular disease, and meningitis. Bauer and Belt cite a patient who regularly and reproducibly responded to the ingestion of Madeira wine with a rise in blood pressure from 160/90 to 280/140. Sudden blood pressure increases have been associated with lead poisoning.

Clinical Features

Generalized neurofibromatosis has been a concurrent finding in about five per cent of patients with pheochromocytomas, its presence in a patient with either intermittent or persistent hypertension makes a search for a chromaffin tumor mandatory

In patients with the adrenal-sympathetic syndrome, production of an attack by massage of a palpable tumor *is practically diagnostic. Such a*

combination of findings, however, has been encountered in less than five per cent of the published cases. In a slightly greater percentage, pressure on the renal and abdominal areas, in the absence of a palpable mass, has caused sudden blood pressure elevation with symptoms similar to those present during a spontaneous paroxysm. It is worthy of mention that chromaffin tumors can be appreciated on physical examination in less than 15 per cent of instances.

In contrast to the suspicion of chromaffin tumor which prevails when paroxysms of hypertension are recorded, the diagnosis is too often overlooked in patients with persistent hypertension. This is particularly true when the variations in the level of arterial pressure are no greater than are commonly observed in essential hypertension. There are several findings which may be observed in this group of patients which make the specific exclusion of a pheochromocytoma more imperative.

In many instances a careful history will disclose episodes which suggest the sudden outpouring of pressor catechols. Sudden bouts of precordial heaviness and palpitation, of pounding fullness in the head, of inexplicable anxiety and tremulousness, and episodes of rather abrupt blanching and mottling of the extremities should be sought for by deliberate questioning

The frequency of excessive sweating has been emphasized. The sweating may be episodic or continuous. In my experience, it has been almost universally present when sought. It should be emphasized that this sweating is more generalized than the palmar, plantar, and axillary hyperhidrosis so often seen with nervous tension.

The hypermetabolism shown by many patients with pheochromocytoma has been discussed earlier. It is not accompanied by enlargement of the thyroid gland, nor by an elevation in serum protein-bound iodine, nor by an augmented avidity of the thyroid for tracer doses of radio-iodine The concurrence of hypertension and hypermetabolism should always raise the question of a chromaffin tumor. It is probable that an increase in the level of circulating epinephrine may mimic the increased sensitivity to epinephrine seen in hyperthyroidism and explain the considerable number of patients with pheochromocytoma who have been mistaken for thyrotoxicosis by clinicians *thoroughly familiar with the latter. Although* statistically hyperglycemia and hypertension have a much less impressive correlation with pheochromocytoma, they should probably be regarded in a similar light.

Smithwick's group was impressed by the fact that many of their patients with hypertension due to pheochromocytoma were hyporeactors to the cold pressor test, whereas a very high percentage of other hypertensives studied by them were hyperreactors Another group in the same city, Evans et al., working with an apparently similar patient population,

found that half of their patients with essential and malignant hypertension were hyporeactors, and all three of their patients with pheochromocytomas were hyperreactors

Smithwick and his colleagues also observed that postural tachycardia and postural hypotension, which were relatively infrequent in essential and malignant hypertension, were present in about half of their patients with chromaffin tumors; other clinics have not reported on these phenomena.

Radiography

In this disease radiography has had its greatest application in the location and confirmation of neoplasms, and has been the technique which first suggested the correct diagnosis in a number of instances. The intrathoracic tumors have been readily found, and plain films of the abdomen have occasionally shown suggestive soft tissue shadows in the adrenal areas, or displacement of the shadow of either a kidney or a gas-filled viscus. Somewhat more often, changes in renal shape or position as demonstrated by intravenous or retrograde pyelography have impelled the roentgenologist to raise the question of a mass in the perirenal area. It is now accepted practice to perform an intravenous pyelogram prior to considering a lumbodorsal sympathectomy, and the tumor in the patient described by Thorn et al., as well as several others, have been so identified

Pheochromocytomas are not the most common causes of renal displacement or distortion When the roentgenologist suggests the possibility of an adrenal or periadrenal tumor, the clinical picture must be re-examined with both cortical and medullary neoplasms in mind, as well as renal, pancreatic, and retroperitoneal new growths. Metabolic studies, assays of urinary steroids, and pharmacologic tests for pheochromocytoma should be appraised in terms of their relevance to the syndrome presented by the patient. If the clinical, radiological, or pharmacological evidence is sufficiently suggestive, more detailed x-ray study of the perirenal areas is facilitated by the technique of perirenal air injection; this has been discussed in detail in a number of publications by Cahill, and by Mencher In many clinics perirenal air insufflation has been regarded as highly hazardous, and a number of surgeons have preferred to do bilateral adrenal explorations rather than employ it In Cahill's hands, untoward reactions have been extremely rare and much valuable information had been gained

As is true of all diagnostic methods for pheochromocytoma, insufflation of gas into the perirenal fascial planes has its limitations. In many instances, even when insufflation is carried out with maximal skill, visualization of the region is inadequate for interpretation Those pheochromocytomas—more than 15 per cent—which lie outside of the perirenal areas will not

be revealed. Finally, the films may show highly suggestive shadows which operation reveals to be due to causes other than adrenal neoplasms. I have had the privilege of reviewing the records of three patients who were explored during the last year because of suspicious histories and shadows which on perirenal aerograms strongly suggested tumors The pharmacological tests were negative in all; one patient had lipoma in the perirenal fat, in another the shadow was produced by the tip of an unusually placed spleen, and in the third the tail of the pancreas was so curled upon itself as to give a golf-ball-like shadow on the aerogram.

Pharmacological Tests

Probably the ideal diagnostic method of separating patients with hypertension due to pheochromocytoma from most other patients with elevated blood pressure would be the identification of excessive quantities of epinephrine and/or norepinephrine in the blood during the episode of hypertension. This has been sought by many, and claimed by not a few. Beer, King, and Prinzmetal were the first to report this finding; they used a modification of the Pissemski method of perfusion of plasma through the arteries of the denervated ear of a rabbit A constant inflow pump was used, and the infusion pressure measured with a manometer The plasma drawn from the patient during an attack caused a rise in perfusion pressure; this rise could be prevented by the prior infusion through the ear of a 1:300,000 solution of ergotamine tartrate. Plasma drawn from a normal control did not cause this rise Hyman and Mencher reported a similar response with plasma drawn from another patient during a paroxysm. Plasma drawn by others under apparently identical conditions has failed to produce a distinctive reaction, and Kreuzfeldt stated that the method itself is non-specific and valueless for quantative comparisons. Strombeck and Hedberg employed a chemical method which depended on the decolorization of methylene blue, they reported that a patient with a pheochromocytoma had a markedly elevated blood level of a substance which reacted like epinephrine in this determination.

The search for a circulating pressor principle in hypertensive vascular disease has led to an enormous number of researches on the blood that have shown that the plasma and whole blood contain many and varied substances which may cause vasoconstriction under experimental conditions.

When pheochromocytomas were found to contain both norephinephrine and epinephrine, specific methods had to be devised which would identify and quantitate each. Workers in Gaddum's laboratory have demonstrated that the biological identification and quantitative assay of these two catechols requires quite a battery of test objects.

Increasingly precise chemical means of separation and estimation have also been developed. James published a chromatographic method which was used by Goldenberg and coworkers in identifying norepinephrine in "U.S.P. epinephrine", and in confirming Holton's finding that chromaffin tumors contained both of these catechols. Von Euler and Hamberg have devised a colorimetric method which depends on the fact that the susceptibility of epinephrine and norepinephrine to oxidation by iodine depends on the pH These two methods have been very useful in the study of tumor extracts, but do not possess sufficient sensitivity to permit their application to circulating blood Lund has published a technique in which the catechols are absorbed from the blood by alumina; epinephrine is then oxidized to adrenochrome, which is converted into the fluorescent compound, adrenolutine He states that this method will detect epinephrine in concentrations of only ten millimicrograms per milliliter.

Engel and von Euler found increased urinary output of epinephrine and norepinephrine in two patients with pheochromocytomas, but their colorimetric method cannot be applied to urines containing pressor amines in only moderately greater than normal quantities.

These techniques have been reviewed in detail by Goldenberg in his recent article on adrenal medullary function In his opinion none is suitable for routine screening use. The absorption feature of Lund's technique has proved technically troublesome, and reproducible results have been hard to obtain The urinary analyses have proved laborious and time-consuming. At the time of writing, chemical and biological assays of urine and blood for increased quantities of catechols seem practicable only when clinical evidence and/or the tests described below make the presence of a pheochromocytoma reasonably probable

There are data which suggest that persistent hypertension due to pheochromocytoma may be maintained for significant periods of time during which there is no excessive level of circulating epinephrine or norepinephrine Under such circumstances the determination of blood catechol levels will have the same weakness as has been demonstrated for the drugs with adrenergic blocking action The value of urinary assays in this situation remains to be determined.

The major differential diagnostic problem presented by patients with paroxysmal elevations of blood pressure is to distinguish those caused by pheochromocytomas from those secondary to diencephalic discharges. This problem remains whether the paroxysms are superimposed on a previously elevated arterial tension, or rise from normal levels

Three pharmacological tests are now available which are said to provoke vasopressor discharges from pheochromocytomas. The diagnostic usefulness of these procedures depends on the production of a greater blood pressure

increase in the presence of a chromaffin tumor than in its absence. All "provocative tests," whether they involve the injection of some drug or merely massage of a palpable tumor or pressure on the abdomen, have hazards. Cardiac arrhythmias, myocardial anoxia, acute left-sided heart failure, and cerebral vascular accidents may follow any spontaneous or induced attack. At least three patients have died during spontaneous paroxysms which have occurred in hospital while they were awaiting operation.

The pharmacological bases of the tests for pheochromocytoma have recently been examined by Entwisle et al. Their findings are not in accord with the suggestion that any drug which causes a drop in blood pressure will cause the discharge of pressor amines by a pheochromocytoma. In addition, a number of patients with chromaffin tumors who have responded positively to the provocative pharmacological tests described below have not shown paroxysms of hypertension when their blood pressures were suddenly and significantly lowered by amyl nitrite or nitroglycerine.

HISTAMINE TEST

The oldest and most widely used of the provocative tests is the histamine test proposed by Roth and Kvale. They first gave histamine intravenously to a patient with pheochromocytoma with the hope that this drug could be used later during operation to counteract the alarming rise in pressure which so frequently occurs Each time histamine was exhibited, even in minute doses, an attack identical with the patient's spontaneous attacks was induced This observation prompted a study of the responses of a larger group of patients to intravenous histamine It was soon observed that a number of patients had rises in blood pressure following the injection of histamine, but in the original 51 patients reported, the pressor response to the injection of histamine exceeded the rise that followed the cold pressor test in only three patients, each of these was found to have a pheochromocytoma.

Technique. The test is easy to perform The patient lies comfortably in the supine position, and the blood pressure is repeatedly determined until a relatively constant base-line is obtained From 0 025 to 0 05 *milligrams* of histamine base is then injected rapidly into the antecubital vein, and the blood pressure followed at 30-second intervals for five minutes and at one-minute intervals for another ten minutes The test is considered positive if histamine produces a greater increase in blood pressure than is caused by the standard cold pressor test in the same patient

Feldberg and Minz demonstrated that the injection of histamine into the adrenal artery of an experimental animal caused adrenal medullary discharge, whereas intravenous injection of similar amounts did not. Ent-

wisle and his colleagues confirmed this finding; they found that the response was not altered by acute denervation of the adrenal gland, and thus excluded the possibility that it was reflexly mediated. There is much to suggest that pheochromocytomas have an increased sensitivity to certain pharmacological agents analogous to the heightened responsiveness to sympathin shown by structures after their postganglionic sympathetic innervation has been destroyed. This would explain the activation of pheochromocytomas by amounts of histamine too small to stimulate the innervated medulla.

The histamine test has been of real value and has been positive in well over 30 patients with proved pheochromocytomas. It has, however, several shortcomings. The most important of these is the rather frequent occurrence of false positive responses; i e., a greater rise in blood pressure after histamine than after the cold pressor test in a patient who does not have a chromaffin neoplasm. All workers who have used the histamine test on a large number of patients have commented on this feature, and the absence of a pheochromocytoma has been confirmed in many by operative exploration. These false positives are usually seen in patients with labile vasomotor systems and other evidences of autonomic imbalance. Since it is in such individuals that the need to differentiate attacks due to pheochromocytoma from anxiety attacks is most likely to arise, this disadvantage is a real one. False negatives have been infrequent but unequivocal; whether or not these depend on some difference in tumor innervation cannot be decided from available data.

ETAMON TEST

LaDue, Murison, and Pack observed a paroxysmal increase in blood pressure after the injection of tetraethylammonium bromide in a patient with a pheochromocytoma who had previously reacted positively to the histamine test They found that this rise could be lessened by tilting the patient from the supine to the upright position When the patient's tumor had been removed, the same procedure did not produce a pressor response.

Technique. With the patient in the recumbent position, the base-line blood pressure is determined. 0.4 grams of tetraethlammonium bromide is then injected intravenously and the blood pressure determined at 30-second intervals for five minutes, and at one-minute intervals for 10 minutes thereafter It has been my practice to consider as positive only those pressor responses which exceeded those produced by the cold pressor test.

Evans and his coworkers reviewed the results of 218 "Etamon Tests" They reported nine "false positive responses" in which increases in arterial pressure greater than those produced by the cold pressor test followed the injection of tetraethyl ammonium ion in standard dosage. Absence of a

pressor response to this drug in the presence of a pheochromocytoma was reported in two instances by Roth and Kvale; another false negative is included in the paper by Beyer et al., and others have been published. Console, Dunbar, and Ray were alarmed by the arrhythmias, tachycardia, and alternating hyper- and hypotension which occurred during a paroxysm induced by Etamon, and suggested that these undesirable effects may be due to interference with autonomic buffering reactions by the ganglionic blocking action of the drug

Intra-arterial injection of tetraethylammonium ion causes a stimulation of the denervated adrenal medulla, and it is probable that the response of pheochromocytomas to this agent is analogous to their response to histamine.

MECHOLYL TEST

Mayock and Rose reported a patient with paroxysms of hypertension due to pheochromocytoma in whom an attack was produced by the subcutaneous injection of ten milligrams of acetyl-β-methyl choline (Mecholyl). The administration of amyl nitrite in amounts sufficient to produce a 40 per cent fall in mean blood pressure did not cause a paroxysm in this patient.

Guarneri and Evans were searching for an agent which might obviate some of the hypertensive complications which occur during the surgical removal of chromaffin tumors when they administered 25 milligrams of Mecholyl subcutaneously to their patient The injection was followed by a paroxysmal increase in blood pressure not dissimilar to that which the patient had after the histamine test It was found that this pressor response to Mecholyl was not abolished by the prior administration of atropine, and that it disappeared when the pheochromocytoma was extirpated. More recently the dosage recommended for the "Mecholyl Test" has been decreased to 10 milligrams, and the incidence of side reactions has been greatly lessened.

Technique With the patient comfortable in the supine position, the baseline blood pressure is determined 0.01 grams of Mecholyl is then injected *subcutaneously* and the blood pressure observed at one minute intervals for half an hour Pressor responses are compared with the cold pressor test responses and only those which exceed them are considered to be positive.

The pharmacological studies of Entwisle et al indicated that Mecholyl had a much less consistent stimulant action on the denervated adrenal medulla when administered intraarterially than did histamine or tetraethylammonion ion.

Save for the studies of Evans and his colleagues, large experiences with the Mecholyl Test have not been published. Several false negatives have

been observed in patients with operatively verified tumors, and the patient studied by Shapiro et al. reacted negatively both to Mecholyl and histamine

Diminution in sensitivity to subcutaneously administered epinephrine was noted preoperatively by Mayock and Rose, and a return to normal responsiveness was observed after the removal of the pheochromocytoma. It seems unlikely that this observation will be of diagnostic value since the rate of absorption of hypodermically injected epinephrine is highly irregular, and the patient's normal sensitivity is usually not established until after the neoplasm has been removed. Furthermore, Hyman and Mencher report that paroxysms may be produced in some patients by epinephrine so given.

Since each of the provocative tests discussed above has its limitations, it is often wise to employ several in puzzling patients, and to evaluate the combined data along with the results of radiographic studies.

Although the clinical and radiographic data are often of value, there remains a considerable number of patients with hypertension secondary to pheochromocytoma who cannot be differentiated from patients with the more common types of blood pressure elevation Since available therapies for the latter are merely palliative and do not beneficially affect the patient with a functioning chromaffin tumor, early identification of the latter is of the utmost importance Until quantitative estimation of epinephrine and norepinephrine in urine and blood is developed to the point where it can be used as a routine screening procedure, we shall have to place our main reliance for this identification on adrenergic blocking agents.

PIPEROXAN TEST

In 1947 Goldenberg, Snyder and Aranow described a test with piperidylmethylbenzodioxane, a dioxane derivative, now known as piperoxan [illegible]

[illegible] due to pheochromocytoma the intravenous injection of 10 milligrams per square meter of body surface produced a fall in blood pressure In patients with blood pressure elevations of other origin the drug produced either no change or an elevation of arterial tension These authors found that the amount of 933F necessary to abolish hypertension caused by excessive amounts of circulating epinephrine was only about one-twentieth of that required to depress or abolish the reflexes originating in the carotid body and aortic arch; it was only about one-fortieth as great as that which caused peripheral sympatholytic actions. The enormous spread between the adrenergic blocking dose and the peripheral sympatholytic dose suggested that the depressor response produced by piperoxan more specifically

indicated hypertension due to circulating epinephrine than did similar responses to agents in which sympatholytic and adrenergic blocking dosages were more nearly equal.

When the piperoxan test was first published, the work of Melville and of others suggested that the benzodioxanes might be much less effective in blocking the action of norepinephrine than of epinephrine. Goldenberg and Aranow, however, demonstrated in human subjects that the recommended dosage of 0.25 milligrams of piperoxan per kilo of body weight used for the test significantly diminished the hypertension produced by the intravenous infusion of norepinephrine. The more recent studies of Melville confirmed this in experimental animals and underlined some of the pitfalls in the interpretation of the "blocking action" of one pharmacological agent of the effects of another. Melville states, "It is clear from the above results that injection of F933 can produce a depressor response during hypertension caused by the presence of either adrenaline or noradrenaline."

Many data suggest that piperoxan competes with the pressor catechols for prosthetic groupings on the receptor tissues. In general, the larger amount of catechol circulating at the time of the piperoxan injection, the greater the depressor response. When the ratio of pressor amine dosage to piperoxan dosage becomes great enough, however, no fall in blood pressure may be observed.

Positive piperoxan tests have been obtained in patients with maintained hypertension due to pheochromocytomas which, on analysis, have been shown to have more than ninety-seven per cent of their pressor amine content in the form of norepinephrine

It is my opinion that accurate interpretations of the results of piperoxan tests are more difficult when modifications of the original technique are introduced. Before evaluating the test, it seems worthwhile to review the procedure which seems to give the most reproducible results.

Technique. The test should be performed only when elevation of the arterial pressure of significant degree exists. The drug should be injected intravenously through a needle which has been introduced into a convenient arm vein before the base-line blood pressure is determined; the needle is kept patent by a slow drip of either dextrose in water or normal saline. The dosage employed should be either 0 25 milligrams per kilo of body weight or 10 milligrams per square meter of body surface and should be injected over a period of two minutes Blood pressure is determined at 30-second intervals from the start of injection for a period of five minutes and at one-minute intervals thereafter for at least 15 minutes. The overall effect of the injection of piperoxan is appraised by plotting the pressure readings against time in minutes and then determining whether the area

above or below the preinjection base-line is the greater. Brief diminutions in tension probably related to cardioacceleration and flushing produced by the drug do not indicate a positive response if accompanied by a greater area above the preinjection baseline than below it Fewer disturbing symptoms are encountered if the side effects of the drug are explained to the patient prior to the test.

The use of piperoxan in paroxysms of hypertension due to pheochromocytoma yields variable results. With very large outpourings of epinephrine and norepinephrine the blocking action of the drug may not be manifest. This has been observed in a patient who had two depressor responses to piperoxan when her hypertension was of constant degree, but had a paroxysmal rise in the middle of a piperoxan-induced fall during a third test. Following the short paroxysm, the pressure again fell to below preinjection levels, and gradually returned to the base-line. In some instances, paroxysms, both spontaneous and induced, seem to be materially shortened by the injection of piperoxan, in others it is difficult to determine whether any effect has been achieved, since the duration of untreated paroxysms cannot be predetermined.

At the time piperoxan was released for general use more than three years ago, 59 patients with proved pheochromocytomas had positive reactions to the test. These 59 cases were found during the year and a half which elapsed between publication of the test and the distribution of the drug to the general medical public. It is not possible to state how many have been discovered since, but it is presumably as great if not greater. The most fruitful application of the test has been in the routine study of patients with persistently elevated blood pressures Almost all workers who have so employed it in large series have uncovered at least one case of pheochromocytoma unsuspected prior to the performance of the test.

In spite of this impressive record, the piperoxan test is by no means ideal, and its drawbacks are detailed below

In a number of patients, particularly in those with clinical pictures which suggested malignant hypertension, the injection of piperoxan has been followed by pressor reactions of alarming magnitude, often accompanied by tachycardia. Drill, Green and Peterson, Wilkins et al, and Bierman and Partridge have reported reactions of this type In Green and Peterson's patient, the test precipitated an attack of hypertensive encephalopathy identical to those the patient had experienced spontaneously; the attack cleared promptly and left no residua. Pulmonary edema followed a pressor response in one of Bierman and Partridge's patients, and the other probably had a true drug idiosyncrasy since marked depressor and pressor reactions followed only a small fraction of the standard dose.

The employment of the piperoxan test in patients with hypertension who

are on the edge of cardiac failure must be undertaken as a calculated risk, although I have learned of no irreversible damage which has followed it.

Five false negative responses to the piperoxan tests are known to me at the time of writing. In all these patients pheochromocytomas were found at operation; it is probable that our data on this type of response are incomplete. The experiments of Goldenberg and Aranow and the finding of positive responses to piperoxan in patients whose chromaffin tumors contained norepinephrine almost exclusively effectively exclude the possibility that these false negatives are due to some peculiar mixture of catechols secreted by the tumors. It seems most probable that these false negative responses were due to the fact that persistent hypertension may result from the intermittent secretion of catecholamines by chromaffin tumors, and that the tests were performed when there was no excess of these substances in the circulation This is discussed in more detail on the section on the relationship between essential hypertensive vascular disease and the persistent hypertension produced by pheochromocytomas

In the large number of patients studied with piperoxan by Goldenberg and Aranow none with uremia was included Raab had reported that the blood of uremic patients contained an increased concentration of sympathin-like substances, but this source of error was not appreciated until Grimson mentioned it. He noted that some patients with nitrogen retention showed depressor responses not only to piperoxan but to Regitine, and in them Shingleton's method suggested an increase in the blood level of circulating epinephrine. It is probable that the false positive result reported by Taliaferro et al , as well as those by Place, by Soffer, and by others, can be similarly explained The degree of depressor response to the piperoxan test decreased with successive testing in the patient reported by Taliaferro et al., and his blood non-protein nitrogen fell from 76 to 41 mg per cent during his hospital stay. The only false positive response known to the author which cannot be explained on this basis is in the patient with an adenoma of the anterior pituitary reported by Conley and Junkerman; the patient exhibited many of the features of Cushing's syndrome and adrenal cortical hyperplasia was found at autopsy Two piperoxan tests were unequivocally positive, and no pheochromocytoma was present Negative tests have been recorded by me in a number of other patients with Cushing's syndrome.

The small but definite incidence of excessive pressor responses to piperoxan makes worthwhile the search for another adrenergic blocking agent which will not cause undesirable side reactions For routine screening of hypertensive patients, a drug with a less highly specific adrenergic blocking action than that of piperoxan would still be useful, since positive responses could be checked with the piperoxan test; the latter has caused untoward effects only in patients who have not had pheochromocytomas.

REGITINE TEST

Emlet, Grimson, Bell, and Orgain have recently recommended the use of 2-(N-p-tolyl-N-(m-hydroxyphenyl)-aminomethyl)-imidazoline hydrochloride (C-7337 or Regitine) for this purpose. The technique is simple and readily applicable in large scale studies. After determining the base-line blood pressure level, five milligrams of Regitine are injected intramuscularly and the blood pressure is determined each minute thereafter for 20 minutes. Emlet et al. found that this dose produced an average fall of ten millimeters of mercury in the systolic pressure and eight mm. in the diastolic pressure of 62 patients with hypertensive vascular disease tested by them; extreme ranges of response are not specifically mentioned in their paper but are stated to be less than those observed after piperoxan. In patients with pheochromocytomas a much more marked and protracted depressor response was manifest Only a twofold increase in dosage of Regitine is necessary to cause a significant lowering in patients with hypertensive vascular disease, probably by sympatholytic action; this contrasts with the more than twenty-fold increase necessary to produce such action with piperoxan Four patients with positive responses both to Regitine and piperoxan were explored and pheochromocytomas removed from all. If wider experience confirms the sensitivity of the "Regitine Test" and its absence of disturbing side reactions (the sole side reaction mentioned was tachycardia, which was a symptom in only seven patients), it should find a real place in the study of all patients with persistently elevated blood pressures.

DIBENAMINE TEST

Another blocking agent (N,N-Dibenzyl-β-Chloroethyl-Amine) Dibenamine has been proposed as a test for the identification of patients with pheochromocytomas producing hypertension. It is noteworthy that, in the original report on the actions of this compound in man, Hecht and Anderson noted that five milligrams of this agent per kilo of body weight were sufficient to lower the blood pressure in slightly more than one-quarter of normotensive patients. Haimovici and Medinets, as well as Wunsch and his coworkers, found that this dose caused significant depression of arterial pressure in the great majority of patients with hypertension In the dosage studied, therefore, this agent cannot be used to distinguish instances of persistent hypertension due to pheochromocytoma from those due to hypertensive vascular disease Dibenamine's effects are long-lasting; postural hypotension usually persists for many hours after its injection Administration must be meticulously careful, extravasation is often followed by slough, and phlebitis after injection is frequent These disadvantages make the drug unsuitable for routine testing, and of questionable value in patients having persistently elevated blood pressures due to pheochromocytomas.

The long duration of the action of Dibenamine, however, may give it a diagnostic role in the study of patients with frequent paroxysms of hypertension. When paroxysms which have been occurring several times daily are abolished for several days following the injection of Dibenamine, it is highly probable that they are due to a chromaffin tumor.

Therapy

When the diagnosis of pheochromocytoma has been established, surgical extirpation of the neoplasm provides the only therapy. The overall mortality for all operative attempts to remove chromaffin neoplasms is about 20 per cent, but in the last five years this has been considerably reduced Consideration should be given to problems in the management of these patients before, during, and after operation as they have a definite bearing on the outcome.

Anesthesia

The anesthetic problems incident to operative treatment of pheochromocytomas have been analyzed by Apgar and Papper in their recent article They conclude, "A safe method which satisfies most requirements appears to be a tranquil induction with thiopental sodium followed by maintenance with ether through an endotracheal airway " They emphasize that anoxia is the greatest hazard during anesthesia, and that it must be scrupulously avoided

Surgical Approaches

Review of the literature and my own personal experience have favorably impressed me with the trans-abdominal approach This permits careful examination of both adrenals and the entire peri-aortic region. In the hands of those familiar with it, control of the blood supply of the neoplasm has been as effective from this approach as from the lumbar route. The operator can not only exclude the presence of other tumors, but can establish that the opposite adrenal is present and apparently normal This knowledge is important if, as is frequently the case, the presence of the pheochromocytoma makes necessary the removal of the entire involved adrenal

Grimson has recently advocated a new transthoracic, trans-diaphragmatic approach which permits palpation of all of the areas in which tumors have been described, and has been very satisfactory for the excision of neoplasms in the adrenal area. It seems probable that a second operation would be necessary to remove tumors which occur much lower than the adrenals.

High lumbar incision has been used by many, and Richards and Hatch advocate simultaneous exposure of both adrenals by this avenue. Thus

requires two surgical teams and produces a significant amount of operative trauma. If no pheochromocytoma is found, the first stage of a lumbo-dorsal splanchnicectomy can then be performed. The incidence of extra-adrenal tumors in the author's cases has been so high that he would hesitate to have a lumbo-dorsal sympathectomy done on a patient with a preoperative diagnosis of pheochromocytoma until these had been completely ruled out.

Operative Complications

There are two major types of operative complications. The first is caused by acute left-sided heart failure, and follows a sharp increase in arterial pressure or the appearance of an acute arrhythmia. The second appears to be caused by the sudden withdrawal from the circulation of the large amounts of pressor catechols to which the body has become adapted.

There has been little dispute about the existence of the first type of complication. Often vascular collapse has followed the rise in blood pressure incident to manipulation of the tumor even when the vascular connections of the latter have not been interrupted. Cardiac asthma or pulmonary edema has been observed in some instances, and the inference seems sound that the collapse is the result of sudden cardiac failure secondary to the discharge of excessive amounts of epinephrine and norepinephrine. Experience with reactions of this sort has led some authors to state that *all* of the hypotensive reactions, not due to excessive hemorrhage, which have followed extirpation of pheochromocytomas are explicable on the basis of cardiac failure I believe that this extrapolation is not justified and is based on the limited experience of its proponents

Reliable observations have been made during removal of functioning pheochromocytomas which show that the precipitous drop in blood pressure, which at times follows the severance of the last vascular connections of the tumor, is not always preceded by a marked increase in pressure nor accompanied by a cardiac arrhythmia In instances of this sort, vasopressor amines are often of dramatic value, this contrasts with their relative inefficacy in combatting the hypotension due to acute cardiac insufficiency. These data support the hypothesis that certain cases of shock following the extirpation of chromaffin tumors are due to a sudden diminution in the concentration of catecholamines in the blood In some cases of this type it has been necessary to infuse pressor amines for many hours. Interruption of the infusion as long as 12 hours after operation has resulted in dangerous decreases in blood pressures which were promptly reversed by readministration of the sympathomimetic amine; this is not the sequence of events which would be anticipated were left-sided heart failure the dominant problem

The adrenergic blocking agents have been employed prior to and during operation to prevent the paroxysmal increases in blood pressure and ar-

rhythmias which may follow handling of the pheochromocytoma. Before *discussing the individual agents it is important to indicate that any adrenergic* blockade may cause undesirable depressor reactions in a patient with chromaffin tumor. During such a blockage the action of norepinephrine is merely abolished, but the pressor effects of epinephrine are *reversed*. This means that an infusion of epinephrine after the establishment of adrenergic blockade results in lowering arterial pressure below control levels, although the cardioaccelerator action of epinephrine is not abolished. I have learned of two fatalities in patients to whom adrenergic *blocking agents were administered during the induction of anesthesia* and before the actual operative manipulation of the tumor; in neither instance was a blood pressure rise observed prior to the appearance of shock.

The alarming fall in blood pressure which has followed the ligation of the vascular connections of chromaffin neoplasms has often been controlled and overcome by the administration of one of the sympathomimetic amines. If the surgeon chooses to administer a drug which will block the rise produced by tumor manipulation, he cannot expect an adrenergic agent to counteract the fall which may follow extirpation of the pheochromocytoma. Fatalities have occurred when vain attempts have been made to combat this type of shock with pressor amines. Grimson et al. solved this dilemma by using 0.5 to 1.0 milliliters of Pitressin intravenously; adrenergic blockade does not influence the vasoconstrictor action of this agent.

ADRENERGIC BLOCKING AGENTS

Piperoxan

The brevity of the blocking action of piperoxan should make possible the control of the pressor problems by repeated administration before the tumor is removed, and yet not produce so protracted a blockade that reversal of the post-removal hypertension would be difficult. The successful employment of piperoxan in this role has been reported by several authors, but it has several drawbacks. The amounts of piperoxan which are tolerable to conscious human subjects have a comparatively weak adrenergic blocking action which can be overcome by large amounts of either epinephrine or norepinephrine. The effects of large and frequent doses in anesthetized subjects are not known, but piperoxan not only does not block the cardioaccelerator action of epinephrine but increases the heart rate.

Dibenamine

Dibenamine is one of the most potent and long-acting adrenergic blocking agents available, and a number of authors have advocated its administration both before and during operation. Its very potency introduces hazards in addition to those already mentioned. The recommended dosage of five

to seven milligrams per kilo of body weight may produce a significant interference with the vascular reflexes which are of importance in maintaining arterial pressure in the face of a sudden diminution in circulating blood volume. Such an interference is of more than academic interest during the removal of tumors so vascular and so closely juxtaposed to major blood vessels as are pheochromocytomas

Some of the undesirable effects of epinephrine reversal were evident in one of the patients reported by Cahill and Monteith. This patient's pheochromocytoma contained a catechol mixture of which epinephrine constituted 86 per cent. The fall in blood pressure caused by the preoperative infusion of Dibenamine considerably exceeded that which followed removal of the tumor; the Dibenamine-induced lowering was accompanied by signs of localized cerebral ischemia which did not appear postoperatively.

Bartels and Cattell suggested that Dibenamine be employed during the actual operative procedure, but the slow onset of its action and the irreversible shock shown by the patient reported by Litman and State have made others hesitant so to use it. Cahill and Monteith allowed at least 24 hours to elapse between Dibenamine infusion and operation in order that only a partial blockade might be present during operation. Decker et al. preferred to delay operation until a pressor response to epinephrine demonstrated that the blocking action used to abolish paroxysms during the three days preceding operation, had completely worn off.

Priscoline

Priscoline was used intravenously in 50-milligram doses by Console et al. both prior to the induction of anesthesia and during operation for pheochromocytoma. In their report they indicated that it was not an ideal blocking agent, and Grimson emphasized other drawbacks. I recently reviewed the history of a patient who was given Priscoline preoperatively and during the induction of anesthesia A profound blood pressure fall occurred before the tumor was actually handled, and this was only partially reversed by norepinephrine and neosynephrin Following excision of the pheochromocytoma irreversible shock occurred, and the patient died 52 hours after operation

Regitine

At the time of writing the most promising compound for the production of adrenergic blockade before and during operation is Regitine Grimson, Emlet and Hamblen have reviewed its use in five cases and Iseri et al have published a confirmatory experience In the latter's case, oral doses of 25 milligrams each, repeated at three hour intervals, made it possible to lower the patient's blood pressure to approximately normal levels during a 27-

day preoperative period. During this time there was great improvement in his general condition. When the drug was withdrawn for six hours, the tension climbed toward pretreatment levels Just before operation 25 milligrams were given orally and three milligrams were injected intramuscularly. Handling of the tumor two and one-half hours later caused the pressure to climb to 200/150 which was less than the values recorded at the time of admission to the hospital Another one-half milligram lowered the pressure to 130/80, and there was a further decline to 90/50 after the excision of the tumor. Progressive fall was prevented by the infusion of eight micrograms of norepinephrine per minute.

Grimson has given repeated intravenous injections of one-twelfth to one-sixth of a milligram per kilo of body weight, or five to ten milligram intramuscular doses That this agent is not without the dangers mentioned earlier was demonstrated by a patient whose tumor was recently analyzed by Goldenberg; this patient received Regitine before the induction of anesthesia and died in shock before the operative procedure was well under way.

To combat the hypotension which may follow the sudden withdrawal of circulating catecholamines, a number of pressor drugs have been administered.

PRESSOR AGENTS

Pitressin

Pitressin has a vasoconstrictor action which is not blocked by any of the adrenergic blocking agents, and it may therefore be employed with effect in patients whose responses to sympathomimetic amines have been abolished. Pitressin, however, is an unphysiological pressor agent and causes constriction of the coronary arteries and general capillary constriction. In a patient with coronary artery diseases or cardiac insufficiency, this action is undesirable It has been injected intravenously by Grimson et al. in doses ranging from 0 5 to 1 0 milliliters

Sympathomimetic Amines

A variety of sympathomimetic amines has been injected to prevent the shocklike state which may follow removal of the tumor Their ineffectiveness in the presence of adrenergic blockade has been indicated. In patients in whom no block exists, or in whom only partial blockade has been established, a continuous infusion of norepinephrine provides the most precise and readily adjustable control of arterial pressure. This catechol does not cause coronary constriction, yet narrows all other important vascular beds. Unlike epinephrine, its action will not be reversed by any partial blockade

which may be present. Norepinephrine is so rapidly metabolized that immediate rises and falls of blood pressure follow changes in the rate of infusion, and its cardioaccelerator action is slight. A solution containing eight micrograms of the racemic compound or four micrograms of l-norepinephrine per milliliter provides adequate flexibility. Neosynephrine closely resembles norepinephrine but is less rapidly metabolized and therefore has a more prolonged action. The relative disadvantages of epinephrine are implicit in the above discussion; it has no advantages over norepinephrine.

CORTICAL PRINCIPLES

When only one of the adrenal glands is involved by the pheochromocytoma, or when the neoplasm is extra-adrenal, there is little evidence that adrenal cortical insufficiency is a major problem in the postoperative management of patients with chromaffin tumors. A variety of disease processes may lead to the destruction of one adrenal cortex; in the presence of a normal contralateral cortex, no symptoms of cortical insufficiency will appear. The possibility that the patient's only functioning adrenal may be involved by the tumor, or that both adrenals may be compromised by bilateral new growths warrants consideration For these reasons, the prophylactic administration of adrenal cortical principles during the immediate pre- and postoperative periods seems rational. Priestley and his colleagues have recently reported on their experiences with 29 patients subjected to bilateral subtotal adrenalectomy. They found that cortisone in dosages of 200 milligrams per day beginning 48 hours preoperatively and continued for the first two or three postoperative days was ample to prevent evidence of adrenal cortical insufficiency in patients from whom almost all cortical tissue had been removed. A smaller amount should be more than sufficient for prophylactic use in patients with chromaffin neoplasms.

Relationships of Persistent Hypertension Due to Pheochromocytoma to Essential Hypertensive Vascular Disease

The practical identity of the clinical picture in patients with essential hypertensive vascular disease and in some patients with pheochromocytoma has been the subject of comment for many years. It was difficult, however, to understand how this picture could be produced by excessive amounts of circulating epinephrine In a number of such cases of pheochromocytoma there was no hyperglycemia or tachycardia at the time of study, and the metabolic rates did not differ significantly from those in essential hypertension

When it was found that norepinephrine might constitute a major fraction of the pressor principle of chromaffin tumors, it seemed that it might be possible to correlate the percentage of the primary amine in the tumor with the closeness with which the syndrome of essential hypertension was mimicked. At first, when only a few tumors had been analyzed, the correlation was fairly good, but widening experience revealed an increasing number of exceptions. On the basis of chemical studies of the pheochromocytomas from 13 patients who exhibited persistent hypertension prior to removal of their tumors, and the clinical data on these patients, Goldenberg et al concluded that tumors containing large total amounts of norepinephrine, and tumors in which epinephrine was the dominant catechol, tended to exhibit striking metabolic features. When the tumor had a small total catechol content which was predominantly norepinephrine, a picture closely resembling essential hypertension was usually seen.

When the first false negative piperoxan test was reported, it seemed probable that this might be caused by the fact that the piperoxan test was not able to detect hypertension produced by excessive amounts circulating norepinephrine Experiments with norepinephrine infusions, however, showed that the usual doses of piperoxan blocked, but did not reverse, the pressor effect of this hormone Shortly thereafter two strikingly positive piperoxan tests were recorded on a patient whose tumor amines were 97 per cent norepinephrine and only three per cent epinephrine Analysis of the tumor of a patient who had shown a false negative piperoxan test revealed a mixture of catechols no different from that found in the tumors of several patients who had reacted positively to piperoxan These observations eliminated the possibility of explaining the false negatives by the nature of the catechol mixture.

It was noted that a patient with persistent hypertension caused by pheochromocytoma had responded positively to piperoxan at one time and negatively at another Another patient with a false negative piperoxan test was found to have a basal metabolic rate of plus one per cent at the time of the false negative test, several weeks previously her BMR had been plus 64 per cent Since her hypermetabolism was due to the secretion of excessive amounts of catecholamines, it appeared probable that their concentration in the circulating blood varied widely in spite of the fact that her blood pressure was constantly elevated.

The pre- and postoperative courses of 12 patients whose tumors were chemically analyzed were then re-examined. In seven of these 12 patients it was noted that hypertension persisted for varying periods of time after the excision of the pheochromocytoma That this was not due to concomitant hypertensive vascular disease was demonstrated by the fact that completely normotensive levels were reached in at least two of the seven

patients after a lapse of several weeks. McCoy and Bridgeman report a 13-year-old boy with persistent arterial hypertension prior to the removal of a pheochromocytoma whose blood pressure was still 150/110 on the 21st postoperative day, and did not reach normal levels until eight months had passed.

These data lead to the conclusion that the prolonged presence of excessive amounts of either epinephrine or norepinephrine in the circulation may lead to vascular readjustment which maintain blood pressure elevation for varying and possibly for indefinite periods of time in their absence. They imply that persistent hypertension may result from only intermittent hypersecretion of epinephrine and norepinephrine. One must also infer that any test for pheochromocytoma based on the detection of excessive amounts of circulating pressor catechols will be handicapped by the same percentage of false negatives; fortunately, this percentage seems to be small

Since the turn of the century when it was found that many of the effects of sympathetic nerve stimulation could be reproduced by the injection of epinephrine, a great deal of research has been directed toward the identification of the chemical transmitter of sympathetic nerve impulses In 1933 Cannon and Rosenblueth proposed the hypothesis that there were two such mediators, which they called "Sympathin E" and "Sympathin I." In the following year Bacq suggested that norepinephrine might be Sympathin E Within the last few years von Euler demonstrated the presence of norepinephrine in mammalian sympathetic nerve fibers He suggested calling norepinephrine "Sympathin N," and epinephrine "Sympathin A," since he believed that varying mixtures of these catechols could reproduce all of the effects of sympathetic nerve stimulation Peart was able to show that norepinephrine was actually released when the sympathetic nerves to the spleen were stimulated At present, there is no final agreement on the chemical nature of sympathin, and the evidence has been examined in detail in Rosenblueth's recent monograph. There is universal agreement, however, that norepinephrine and epinephrine bear a very intimate relationship to the transmitting substance or substances, if they are not actually identical The importance of the sympathetic innervation of the vascular tree in the maintenance of arterial pressure needs no emphasis It seems certain that whatever the role of psychoemotional tensions in the production and maintenance of hypertension, these effects are achieved over sympathetic nervous pathways The surgical interruption of a considerable number of such pathways is the most widely accepted therapy for hypertensive vascular disease.

Studies of patients with essential hypertension have revealed no more than normal amounts of epinephrine and norepinephrine in the blood or urine It is upon this fact that the detection of hypertension due to pheo-

chromocytoma by means of adrenergic blocking agents depends. The hypertensive state which persists in some patients after the surgical removal of the chromaffin tumor which provoked it seems in all ways identical with hypertensive vascular disease. Pre-operative hypermetabolism and hyperglycemia disappear, the responses to the adrenergic blocking agents become normal, and there is no evidence of excessive secretion of epinephrine or norepinephrine

The persistence of this hypertension after the withdrawal from the circulation of the excess of catecholamines which provoked it suggests an analogy with the findings of Farris et al. on the persistence of experimental audiogenic hypertension after the acoustic stimulus has ceased.

Do these findings imply that sympathin-mediated hypertension—i e, hypertension due either to excessive amounts of circulating sympathin as in pheochromocytoma, or to excessive amounts of sympathin released at myoneural junctions as in neurogenic hypertension—has a special relationship to hypertensive vascular disease? Or will any blood pressure elevation, if frequent and prolonged enough, produce a hypertension which will outlive the stimulus which caused it?

The answer to these questions should greatly broaden our knowledge of the entire problem of hypertension, and it seems worthwhile to turn the battery of our investigative techniques on those patients with pheochromocytoma in whom an elevated blood pressure persists after operation. Patients with coincidental hypertensive vascular disease can be excluded by restricting our analysis to the data from those patients whose blood pressures eventually reach normal levels. Such studies may indicate that the crucial problem in hypertensive vascular disease is not what causes the initial elevation of pressure, but what perpetuates it when the primary stimulus has ceased

SUMMARY

The clinical picture presented by patients with pheochromocytomas has been analyzed in the light of our knowledge of the hormones elaborated by them—norepinephrine and epinephrine.

The differential diagnosis of pheochromocytoma has been discussed. It is emphasized that the major problem is to distinguish those patients with persistent hypertension secondary to chromaffin tumors from the great number of patients with essential hypertensive vascular disease The clinical features and pharmacological tests which are useful in making this distinction have been described and evaluated

Tests of value in distinguishing paroxysmal hypertension due to pheochromocytoma from other causes of episodic blood pressure elevation have been detailed, and their merits and shortcomings weighed

Some of the problems of operative management of patients with chromaf-

fin tumors have been analyzed, and views on optimal methods of handling them presented.

Finally, some of the provocative similarities and possible relationships of the persistent hypertension due to pheochromocytoma to essential hypertensive vascular disease have been indicated.

BIBLIOGRAPHY

1 Apgar, V. and Papper, E. M. Pheochromocytoma: Anesthetic management during surgical treatment Arch Surg, 62: 634, 1951

2 Aranow, H, Jr Differential diagnosis of pheochromocytoma M Clin. North Am, 34: 757, 1950.

3. Aranow, H, Jr. Pheochromocytoma. In: Progress in Clinical Endocrinology, ed. by Soskin, S. Grune and Stratton, N. Y C, 1950.

4. Bacq, Z. M. La pharmacologie du système nerveux autonome et particulièrement du sympathique d'après la théorie neurohumorale. Ann physiol, 10: 467, 1934

5 Bacq, Z. M. et Fredericq, H Action adrenolytique d'un derive du dioxane (933F). Compt rend Soc biol, 117: 806, 1934

6 Barcroft, H. and Konzett, H. On the actions of noradrenaline, adrenaline, and isopropyl-noradrenaline on the arterial blood pressure, heart rate, and muscle blood flow in man J. Physiol, 110: 194, 1949

7. Barnett, A J, Blacket, R. B, Depoorter, A. E, Sanderson, P H. and Wilson, G M The action of noradrenaline in man and its relation to phaeochromocytoma and hypertension. Clin. Sci, 9: 151, 1950

8 Bartels, E C and Arnold, W T Essential features for diagnosis of pheochromocytoma. report of a case Lahey Clin. Bull, 6: 132, 1949

9 Bartels, E C and Cattell, R B Pheochromocytoma its diagnosis and treatment Ann. Surg, 131: 903, 1950

10 Bartels, E C and Wall, N M Clinical problem of pheochromocytoma Surg Clin North Am, 27: 605, 1947

11 Bauer, J and Belt, E Paroxysmal hypertension with concomitant swelling of thyroid due to pheochromocytoma of right adrenal gland cure by surgical removal of pheochromocytoma J Clin Endocrinol, 7: 30, 1947

12. Bayer, O, Blumberger, K and Efforr, S Cardiovascular effects of norepinephrine in man Cardiologia, 16: 145, 1950

13 Becker, M C., Bass, R. D and Robbins, C M Pheochromocytoma a cause of post-partum convulsions J Med Soc N J, 46: 339, 1949

14 Becker, M C, Bass, R D and Robbins, C M Pheochromocytoma diagnosis and treatment Postgrad Med, 6: 408, 1949

15 Beer, E., King, F H and Prinzmetal, M Pheochromocytoma with demonstration of pressor (adrenalin) substance in the blood preoperatively during hypertensive crises Ann Surg, 106: 85, 1937

16 Belt, A E and Powell, T O Clinical manifestation of chromaffin cell tumors arising from the suprarenal medulla (suprarenal sympathetic syndrome) Surg, Gyn. & Ob, 59: 9, 1934

17 Bernal, P Crises Hypertensives E Goin et Cie, Paris, 1933

18. Beyer, K H, Ross, C A, Wiebelhaus, V D., Waller, W S and Schuchardt, G S Vasopressor components of pheochromocytoma Ann Int Med, 35: 117, 1951

chromocytoma by means of adrenergic blocking agents depends. The hypertensive state which persists in some patients after the surgical removal of the chromaffin tumor which provoked it seems in all ways identical with hypertensive vascular disease. Pre-operative hypermetabolism and hyperglycemia disappear, the responses to the adrenergic blocking agents become normal, and there is no evidence of excessive secretion of epinephrine or norepinephrine.

The persistence of this hypertension after the withdrawal from the circulation of the excess of catecholamines which provoked it suggests an analogy with the findings of Farris et al. on the persistence of experimental audiogenic hypertension after the acoustic stimulus has ceased.

Do these findings imply that sympathin-mediated hypertension—i.e., hypertension due either to excessive amounts of circulating sympathin as in pheochromocytoma, or to excessive amounts of sympathin released at myoneural junctions as in neurogenic hypertension—has a special relationship to hypertensive vascular disease? Or will any blood pressure elevation, if frequent and prolonged enough, produce a hypertension which will outlive the stimulus which caused it?

The answer to these questions should greatly broaden our knowledge of the entire problem of hypertension, and it seems worthwhile to turn the battery of our investigative techniques on those patients with pheochromocytoma in whom an elevated blood pressure persists after operation. Patients with coincidental hypertensive vascular disease can be excluded by restricting our analysis to the data from those patients whose blood pressures eventually reach normal levels. Such studies may indicate that the crucial problem in hypertensive vascular disease is not what causes the initial elevation of pressure, but what perpetuates it when the primary stimulus has ceased.

Summary

The clinical picture presented by patients with pheochromocytomas has been analyzed in the light of our knowledge of the hormones elaborated by them—norepinephrine and epinephrine.

The differential diagnosis of pheochromocytoma has been discussed. It is emphasized that the major problem is to distinguish those patients with persistent hypertension secondary to chromaffin tumors from the great number of patients with essential hypertensive vascular disease. The clinical features and pharmacological tests which are useful in making this distinction have been described and evaluated.

Tests of value in distinguishing paroxysmal hypertension due to pheochromocytoma from other causes of episodic blood pressure elevation have been detailed, and their merits and shortcomings weighed.

Some of the problems of operative management of patients with chromaf-

41. Colston, J. A. C. Surgical aspects of bilateral familial pheochromocytoma. Tr. Am. Assoc Genito-Urin. Surg, 39: 81, 1947.
42 Conley, J. E and Junkerman, C. L. Lack of specificity of piperoxan hydrochloride test for adrenal medullary tumors J. A. M. A., 147: 921, 1951.
43 Consolf, A. D, Dunbar, H. S and Ray, B. S. Pheochromocytoma the use of adrenergic blocking agents in the operative management. Surgery, 28: 428, 1950.
44. Cross, G. O. and Pace, J. W. Malignant pheochromocytoma with paroxysmal hypertension and metastasis to the cervical spine J. A. M A., 142: 1068, 1950
45 Dana, G. W. and Calkins, E. Clinical experience with benzodioxane. Bull J. Hopk. Hosp, 84: 283, 1949.
46 Davis, F. W., Jr, Hull, J. G. and Vardell, J. C., Jr. Pheochromocytoma with neurofibromatosis Am J. Med, 8: 131, 1950
47. Decker, H. C, McDowell, F. W. and Trimble, I. R. Pheochromocytoma: case report with discussion of differential diagnosis and surgical treatment. J. A. M. A, 147: 642, 1951.
48. de Largy, C, Greenfield, H. D. M., McCarry, R. L and Whelan, R F The effect of intravenous infusion of mixtures of l-adrenaline and l-noradrenaline on the human subject Clin. Sci, 9: 71, 1950
49 de Vleeschouwer, G Au sujet de l'action du diethylaminomethyl-3-benzodioxane (F883) et du piperido-methyl-3-benzodioxane (F933) sur le système circulatorie Arch. Internat. de pharmacodyn et de thérap, 50: 251, 1935
50 de Vries, A, Rachmilewitz, M. and Schumert, M. Pheochromocytoma with diabetes and hypertension. Am. J. Med, 6: 51, 1949
51 Drill, V. A. Reactions from the use of benzodioxane (933F) in diagnosis of pheochromocytoma. New Eng J Med, 241: 777, 1949
52 Duncan, L. E, Jr, Semans, J. H. and Howard, J. E Adrenal medullary tumor (pheochromocytoma) and diabetes mellitus: disappearance of diabetes after removal of the tumor. Ann Int Med., 20: 815, 1944.
53 Emlet, J. R., Grimson, K. S, Bell, D M. and Orgain, E S Use of piperoxan and regitine as routine tests in patients with hypertension J A. M A, 146: 1383, 1951.
54 Engel, A. and von Euler, U. S Diagnostic value of increased urinary output of noradrenaline and adrenaline in pheochromocytoma Lancet, 2: 387, 1950
55. Engel, E L, Mencher, W H. and Engel, G L "Epinephrine shock" as manifestation of pheochromocytoma of adrenal medulla report of case with successful removal of tumor. Am. J Med Sci, 204: 649, 1942
56 Entwisle, G, Stone, C. A and Loew, E R Pharmacologic basis of various tests used in the diagnosis of pheochromocytoma Am J Med, 11: 461, 1951.
57 Espersen, T. and Dahl-Iversen, E Clinical picture and treatment of pheochromocytomas of suprarenal two own cases, one with paroxysmal hypertension improved by treatment with methyl thiouracil and cured by surgical intervention Acta Chir Scandinav, 94: 271, 1946
58 Espersen, T and Jorgensen, J Electrocardiographic changes in paroxysmal hypertension due to chromaffin adrenal tumor Acta Med Scand, 127: 494, 1947.
59 Evans, J A, Rubitsky, H J, Bartels, C C and Bartels, E C Re-evaluation of the reliability of pharmacologic and cold pressor studies in hypertension and pheochromocytoma Am J Med, 11. 448, 1951
60 Farris, E J, Yeakel, E H and Medoff, H S Development of hypertension

19. BIERMAN, H R AND PARTRIDGE, J. W. Untoward reactions to tests for pheochromocytoma New Eng J. Med., 244: 582, 1951.
20 BING, R. J. AND THOMAS, C B Effect of two dioxane derivatives, 883F and 933F, on normal dogs and on animals with neurogenic and renal hypertension J. Pharmacol. & Exp Therap, 83: 21, 1945
21. BISKIND, G R, MEYER, M. A AND BEARDNER, S. A. Adrenal medullary tumor: pheochromocytoma cured by surgical intervention: clinical management, analysis of all reported operated cases J. Clin. Endocrinol, 1: 113, 1941.
22 BOVET, D. AND SIMON, A Recherche sur l'activité sympatholytique de dérivés de l'amino methyl benzodioxane Arch. internat. de pharmacodyn et thérap, 55: 15, 1937
23 BOWEN, G L, GRANDIN, D J, JULIEN, E. E AND KRECH, S, JR. Pheochromocytoma complicating pregnancy Am J Ob. & Gyn., 59: 378, 1950
24 BRUCE, G. M The ocular fundus in pheochromocytoma of adrenal gland Tr Am Opth Soc, 45: 201, 1947.
25 BRUNSCHWIG, A Hypertension from pheochromocytoma. J A. M. A, 134: 253, 1947
26 BRUNSCHWIG, A AND HUMPHREYS, E Excision of pheochromocytoma following near fatal attack of paroxysmal hypertension J A. M. A, 115: 355, 1940
27 BURN, J H AND HUTCHEON, D E The action of noradrenaline Brit J Pharm & Chemother, 4: 373, 1949
28 BURRAGE, W C AND HALSTED, J A Adrenal medullary tumor (pheochromocytoma) case report with successful operation. Ann Int Med, 28: 838, 1948
29 CAHILL, G F Air injections to demonstrate adrenals by x-ray J Urol, 34: 238, 1935
30 CAHILL, G F Tumors of the adrenal and the use of air insufflation in their diagnosis Radiology, 37: 533, 1941
31 CAHILL, G F Hormonal tumors of adrenal Surgery, 16: 233, 1944.
32 CAHILL, G F Pheochromocytomas J A M A, 138: 180, 1948
33 CAHILL, G F AND ARANOW, H, JR Pheochromocytoma diagnosis and treatment Ann Int Med, 31: 389, 1949
34 CAHILL, G F AND MELICOW, M M Tumors of the adrenal gland J Urol, 64: 1, 1950
35 CAHILL, G F AND MONTEITH, J C The use of dibenamine and norepinephrine in the operative treatment of pheochromocytoma New England J Med, 244: 657, 1951
36 CALKINS, E, DANA, G W AND HOWARD, J E Current methods of diagnosis of pheochromocytoma J A M A, 145: 880, 1951
37. CALKINS, E, DANA, G W, SEED, J C AND HOWARD, J E On piperidylmethylbenzodioxane (933F), hypertension and pheochromocytoma J. Clin Endocrinol, 10: 1, 1950
38 CALKINS, E AND HOWARD, J E Bilateral familial pheochromocytomata with paroxysmal hypertension successful surgical removal of tumors in 2 cases with discussion of certain diagnostic procedures and physiological considerations J Clin Endocrinol, 7: 475, 1947
39 CANNON, W B AND ROSENBLUETH, A Studies on conditions of activity in endocrine organs XXIX Sympathin E and sympathin I Amer J Physiol, 104: 557, 1933
40 CANNON, W B AND ROSENBLUETH, A Autonomic Neuro-Effector Systems The MacMillan Co, New York, 1937

82. GRIMSON, K. S. Discussion. J. A M A., 145: 883, 1951
83 GRIMSON, K S, EMLET, J. R. AND HAMBLEN, E. C. Diagnosis and management of tumors of the adrenal gland. Ann. Surg, 134: 451, 1951
84 GRIMSON, K S, HENDRIX, J. P. AND REARDON, M J. Newer adrenolytic, sympatholytic, and ganglionic blocking drugs. J A M A., 139: 154, 1949.
85 GRIMSON, K. S, LONGINO, F. H, KERNODLE, C E. AND O'REAR, H B Treatment of a patient with a pheochromocytoma use of an adrenolytic drug before and during operation J. A. M. A, 140: 1173, 1949
86 GUARNERI, V AND EVANS, J. A Pheochromoctyoma, report of a case with a new diagnostic test Am J. Med, 4: 806, 1948
87. HAIMOVICI, H. AND MEDINETS, H. E Effect of dibenamine on blood pressure in normotensive and hypertensive subjects. Proc. Soc. Exper. Biol & Med., 67: 163, 1948
88 HAMILTON, J. E. Pheochromocytoma of adrenal with paroxysmal hypertension case relieved by surgery. Kentucky M. J, 38: 572, 1940
89 HANDSCHIN, E Zur Kenntnis der Zuckerkandlischen Organe Beitr z path Anat u. z allg Path, 79: 728, 1928
90 HATCH, F. N., RICHARDS, V. AND SPIEGL, R. J Adrenal medullary tumor (pheochromocytoma) Am J. Med, 6: 633, 1949
91. HEATH, F K, CAHILL, G. F AND ATCHLEY, D W Pheochromocytoma correct diagnosis and successful operation J A M A, 117: 1258, 1941
92 HEDINGER, E Struma Medullaris cystica suprarenalis (Beitrag zur Lehre der Paraganglione) Frankfurt Ztschr f. Path, 7: 112, 1911
93 HELLY, K Zur Pathologie Nebenniere Munchen med. Wchnschr, 60: 1811, 1913.
94 HERDE, M Zur Lehre der Paraganglione der Nebenniere Arch f Klin. Chir, 97: 937, 1912.
95 HEYMANS, C. AND BOUCKAERT, J J. Au sujet des influences du piperido-methyl-3-benzodioxane (933F) sur le système circulatoire Compt rend Soc biol, 120: 79, 1935
96 HOCH, G F Pheochromocytoma with paroxysmal hypertension J Urol, 61: 473, 1949.
97 HOFBAUER, J. Pregnancy changes in the uterine cervical ganglia (Frankenhauser) as causative factors of vascular hypertension in toxemia Am J. Ob & Gyn, 59: 1383, 1950
98 HOLTON, P Noradrenaline in adrenal medullary tumours Nature, 163: 217, 1949
99 HOLTON, P. Noradrenaline in tumours of the adrenal medulla J Physiol, 108: 525, 1949
100 HOWARD, J E AND BARKER, H. Paroxysmal hypertension and other clinical manifestations associated with benign chromaffin cell tumors (pheochromocytoma) Bull. Johns Hopk. Hosp, 61: 371, 1937
101 HUGGINS, C AND BERGENSTAL, D M Surgery of the adrenals J A M A, 147: 101, 1951
102 HULTZ, P AND SCHUMANN, H J Uber das Vorkommen von Arterenol in den Nebennieren Naturwiss, 35: 191, 1948
103 Hyman, A and Mencher, W H Pheochromocytoma of adrenal gland, J Urol, 49: 755, 1943
104 ISERI, L T, HENDERSON, H W AND DERR, J W Use of adrenolytic drug, Regitine, in pheochromocytoma Am Heart J, 42: 129, 1951

in emotional gray Norway rats after air blasting. Am J. Physiol, 144: 331, 1945.
61. Ferraro, L. R. and Angle, R. G. Pheochromocytoma with symptoms of epinephrine shock. Arch. Int. Med., 81: 793, 1948.
62. Fertig, H. H., Taylor, R. D., Corcoran, A. C. and Page, I. H. The renal manifestations of pheochromocytoma: report of a case. Ann. Int. Med, 35: 1358, 1951.
63. Forsgren, A L, Nesset, W D. and Anderson, D. M. Pheochromocytoma of the adrenal with successful removal: report of a case Minnesota Med, 32: 170, 1949
64. Fourneau, E. and Bovet, D. Recherches sur l'action sympathicolytique d'un nouveau dérivé du dioxane Arch. Internat. de pharmacodyn et de thérap, 46: 178, 1933.
65. Fränkel, F. Ein Fall von doppelseitingem, völlig latent verlaufenen Nebennierentumor und gleichzeitiger Nephritis mit Veränderungen am Circulationsapparat und Retinitis. Arch. f. path Anat. 103: 244, 1886.
66. Freeman, N. E, Freedman, H and Miller, C C. Production of shock, by prolonged continuous injection of adrenalin in unanesthetized dogs. Am J. Physiol, 131: 545, 1941.
67. Fulton, R L Benzodioxane test in hypertension: a case report with autopsy findings Ohio State Med J., 47: 127, 1951
68. Gaddum, J. H Estimation of substances liberated by adrenergic nerves In: Methods in Medical Research, 3: 116, Year Book Publishers, Chicago, 1950
69. Gaddum, J H, Peart, W S and Vogt, M The estimation of adrenaline and allied substances in blood J Physiol, 108: 467, 1949
70. Ganem, E J and Cahill, G F Pheochromocytomas coexisting in adrenal gland and retroperitoneal space with sustained hypertension report of case with surgical care New Eng J Med, 238: 692, 1948.
71. Goldenberg, M Adrenal medullary function Am J Med, 10: 627, 1951.
72. Goldenberg, M, Apgar, V, Dfterling, R A and Pines, K L Norepinephrine as a pressor drug J A M A, 140: 776, 1949
73. Goldenberg, M and Aranow, H, Jr Diagnosis of pheochromocytoma by adrenergic blocking action of benzodioxane J A M A, 143: 1139, 1950
74. Goldenberg, M, Aranow, H, Jr, Smith, A A and Faber, M Pheochromocytoma and essential hypertensive disease Arch Int Med, 86: 823, 1950
75. Goldenberg, M, Faber, M, Alston, E J and Chargaff, E C Evidence for the occurrence of nor-epinephrine in the adrenal medulla Science, 109: 534, 1949
76. Goldenberg, M, Pines, K L, Baldwin, E deF, Greene, D G and Roh, C E Hemodynamic response of man to nor epinephrine and epinephrine and its relation to the problem of hypertension Am J Med, 6: 792, 1948
77. Goldenberg, M, Snyder, C H and Aranow, H, Jr New test for hypertension due to circulating epinephrine J A M A, 135: 971, 1947
78. Goldner, M. G Pheochromocytoma with diabetes case report and discussion J. Clin Endocrinol, 7: 716, 1947
79. Graham, J B. Pheochromocytoma and hypertension collective review Surg, Gyn. & Ob, 92: 105, 1951
80. Green, D. M. Pheochromocytoma and chronic hypertension J A M A, 131: 1260, 1946.
81. Green, D. M and Peterson, E M Hypertensive encephalopathy after administration of benzodioxane J A M A, 142: 408, 1950

124. McCullagh, E. P. and Engel, W. J. Pheochromocytoma with hypermetabolism, report of 2 cases Ann Surg., 116: 61, 1942
125 McFarland, G. E, Jr and Bliss, W. R Hemorrhage from spontaneous rupture of a pheochromocytoma of the right adrenal gland Ann Surg, 133: 404, 1951.
126 McKenzie, D W. and McEachern, D Adrenal pheochromocytoma The syndrome of paroxysmal hypertension Tr Am Assoc. Genito-Urin Surg, 31: 127, 1938.
127. MacKeith, R. Adrenal-sympathetic syndrome chromaffin tissue tumour with paroxysmal hypertension. Brit Heart J., 6: 1, 1944
128. Madison, L L. Comparison of the anterior pituitary adrenal cortical stimulating effect of U.S.P. epinephrine, synthetic l-epinephrine, and nor-epinephrine. J. Clin Invest, 29: 789, 1950
129 Maier, H. Intrathoracic pheochromocytoma with hypertension Ann Surg, 130: 1059, 1949.
130 Masson, P and Martin, J. F. Paragangliome surrénal étude d'un cas humain de tumeurs malignes de la medullo surrénale Bull de l'Assoc franç p l'étude de cancer, 12: 135, 1923.
131 Mayo, C. H Paroxysmal hypertension with tumor of retroperitoneal nerve report of case J A M A, 89: 1047, 1927
132. Maycock, R. L and Rose, E Insensitivity to epinephrine in a patient with a functioning tumor of the adrenal medulla Am J Med Sci., 213: 324, 1947
133 Medoff, H. S. and Bongiovanni, A M Blood pressure in rats subjected to audiogenic stimulation Am J Physiol, 143: 300, 1945
134 Melville, K I. Antisympathomimetic action of dioxane compounds (F883 and F933) with special reference to vascular responses to dihydroxyphenylethanolamine (arterenol) and nerve stimulation J Pharm & Exp Therap, 59: 317, 1937
135 Melville, K I Blood pressure effects of noradrenaline and adrenaline with special reference to their antagonism by ergotoxine and other blocking agents J. Physiol, 113: 346, 1951
136 Mencher, W. H Perirenal insufflation J A M A, 109: 1338, 1937
137 Mencher, W. H Paroxysmal hypertension caused by pheochromocytoma of the adrenal gland J Mt Sinai Hosp, 10: 743, 1944
138 Morison, R S and Lissak, K Observations on mode of action of piperidinomethyl benzodioxane (933F) Am J Physiol, 123: 404, 1938
139 Muntz, H H, Ritchey, J O and Gatch, W D Adrenalin producing tumor (pheochromocytoma) containing 2,300 mg of adrenalin Ann Int Med, 26: 133, 1947.
140 Neff, F. C, Tice, G M, Walker, G A and Orkerblad, N Adrenal tumor in a female infant with hypertrichosis, hypertension, overdevelopment of external genitalia, obesity, but absence of breast enlargement J Clin Endocrinol, 2: 125, 1942
141 Nickel, J F, Smythe, C McC and Bradley, S E Effect of pressor agents on renal water and electrolyte excretion in man Fed Proc, 1952, in press
142 Nickerson, M The pharmacology of adrenergic blockade J Pharm & Exp Therap, Part II, 95: 27, 1949
143 Nuzum, F R and Dalton, J W Paroxysmal and persistent hypertension in association with lesions of the adrenal glands Am Heart J, 16: 643, 1938
144 Oberling, C and Jung, G Paragangliome de la surrénale avec hypertension paroxystique Bull et mém Soc méd d'hôp de Paris, 51: 366, 1927

105 James, W O. Demonstration and separation of noradrenaline, adrenaline, and methyladrenaline. Nature, **161**: 851, 1948
106 Kappert, A , Sutton, G C , Reale, A , Skoglund, K. H. and Nylin, G The clinical response of human beings to l noradrenaline and its clinical applicability Acta cardiol , **5**: 121, 1950.
107 King, E S J Malignant phaeochromocytoma of adrenals Path. & Bact , **34**: 447, 1931.
108 Koffler, I , Buck, G , Wingard, C , Hitchcock, P., Guthrie, R. and Teague, R S A case of pheochromocytoma diagnosis by the benzodioxane test, urinary hormone studies, and norepinephrine assay of the tumor. J Clin Endocrinol , **10**: 897, 1950
109. Kohn, A Die Paraganglien Arch f mikro Anat , **62**: 263, 1903
110 Kositchek, R J and Rabwin, M H. Pheochromocytoma successfully removed with the aid of piperoxan (Benodaine) hydrochloride. J. A. M. A , **144**: 826, 1950
111 Kvale, W F , Roth, G M , Clagett, O T and Dockerty, M B Headache and paroxysmal hypertension observations before and after surgical removal of pheochromocytoma Surg Clin North Am , **24**: 922, 1944
112 Kreuzfeldt, H Attempts to demonstrate the presence of pressor substances in the blood of patients with renal hypertension Acta med Scand , **125**: 171, 1946
113 Labbe, M , Tinel, J and Doumer Crises solaries et hypertension paroxystique en rapport avec une tumeur surrénale Bull et mém Soc. med d hôp de Paris, **46**: 982, 1922
114 LaDue, J S , Murison, P J and Pack, G T Use of tetraethylammonium bromide as a diagnostic test for pheomochromocytoma Ann Int Med , **29**: 914, 1948
115 Lands, A M , Nash, V L , Dertinger, B J , Granger, H R and McCarthy, H M The pharmacology of compounds structurally related to hydroxytyramine J Pharm and Exp Therap , **92**: 369, 1948
116 Litman, N N and State, D Pheochromocytoma use of N,N-dibenzyl β-chloroethylamine (dibenamine) and piperidino methyl-benzodioxane (Benzodioxane) in surgical therapy Pediatrics, **4**: 735, 1949
117 Longino, F H , Grimson, K S , Chittum, J R and Metcalf, B H Effects of a new quaternary amine and a new imidazoline derivative on the autonomic nervous system Surgery, **26**: 421, 1949
118 Lu, F C and Melville, K I Effects of noradrenaline on coronary flow and heart contraction as recorded concurrently in the isolated rabbit heart J Physiol , **113**: 365, 1951
119 Luduena, F P , Ananenki, E , Siegmund, O H and Miller, L C Comparative pharmacology of the optical isomers of arterenol J Pharm & Exp Therap , **95**: 155, 1949

121

122

123.

169 SMITHWICK, R R, GREER, W. E. R, ROBERTSON, C. W. AND WILKINS, R W. Pheochromocytoma. a discussion of symptoms, signs, and procedures of diagnostic value. New Eng J Med., 242: 252, 1950
170. SNYDER, C H. AND VICK, E H. Hypertension in children caused by pheochromocytoma: report of 3 cases and review of literature. Am J Dis Child, 73: 581, 1947.
171. SOFFER, A. False positive reaction to the piperoxan hydrochloride test for pheochromocytoma J. A. M A, 148: 538, 1952
172 SOFFER, L., MENCHER, H W. AND COLP, R Pheochromocytoma of the adrenal gland A report of two cases with operative removal of the tumor. Surg Clin North Am, 26: 368, 1946
173 SPAETH, W. AND REVENO, W. Diagnostic tests for pheochromocytoma Harper Hosp Bull, 7: 293, 1949
174 SPATT, S D AND GRAYEL, D. M. Pheochromocytoma of the adrenal medulla A clinicopathological study of five cases Am J. Med. Sci, 216: 39, 1948
175 SPEAR, H C. AND GRISWOLD, D. Use of dibenamine in pheochromocytoma report of a case New Eng J. Med, 239: 736, 1948.
176 SPUHLER, O, WALTHER, H AND BRUNNER, W Zur Diagnose, Klinik, und operativen Therapie des Phaeochromocytoms: Histamintest und Dibenamin Schweiz med Wchnschr, 79: 357, 1949
177. STARR, I, GAMBLE, C. J., MARGOLIES, H. DONAL, J S, JOSEPH, N. AND EAGLE E. Clinical study of the action of ten commonly used drugs on cardiac output, work, and size on respiration, on metabolic rate, and on the electrocardiogram J Clin. Invest, 16: 799, 1937
178 STEHLE, R L AND ELLSWORTH, H C Dihydroxyphenyl ethanolamine (arterenol) as a possible sympathetic hormone J Pharm & Exp Therap, 69: 114, 1937.
179 STRÖMBECK, J. P AND HEDBERG, T P Tumor of suprarenal medulla associated with paroxysmal hypertension Acta chir Scand, 82: 177, 1939
180 SWAN, H J C. Effect of noradrenaline on the human circulation Lancet, 508, 1949
181 TALIAFERRO, I, ADAMS, R A AND HAAG, H B Benzodioxan test fall in pressure following its use in a case of renal hypertension J. A M A, 140: 1271, 1949
182 THORN, G. W, HINDLE, J. A AND SANDMAYER, A Pheochromocytoma of adrenal associated with persistent hypertension case report Ann Int. Med, 21: 122, 1944
183 TULLAR, B F The separation of l-arterenol from natural U S P epinephrine Science, 109: 536, 1949
184 VAN BUCHEM, F S R The hypertensive diencephalic syndrome Acta Med Scand, 130: 575, 1948
185 VAQUEZ, H AND DONZELOT, E Les crises d'hypertension artérielle paroxystique Presse Méd, 34: 1329, 1926
186 VOGT, M Observations on some conditions affecting the rate of hormone output by the suprarenal cortex J Physiol, 103: 317, 1944
187 VON EULER, U S A specific sympathomimetic ergone in adrenergic nerve fibers and its relations to adrenaline and nor adrenaline Acta physiol Scand, 12: 73, 1946
188 VON EULER, U S Identification of the sympathomimetic ergone in adrenergic nerves of cattle (sympathin N) with laevo-noradrenaline Acta physiol Scand, 16: 63, 1948
189 VON EULER, U. S Estimation of adrenaline and noradrenaline in tissue extracts

145 OWENS, F. M., JR Relief of chronic hypertension by excision of a pheochromocytoma. *Arch. Surg*, 59: 896, 1949
146 PAPPER, E. M. AND CAHILL, G. F. Anesthetic problems in hormonal disorders of the adrenal glands. J. A. M A, 148: 174, 1952
147. PAUL, F Die Krankhafte Funktion der Nebenniere und ihr gestaltlicher Ausdruck Virchow's Arch f path. Anat., 282: 256, 1931.
148 PFART, W. S The nature of splenic sympathin J. Physiol., 108: 491, 1949
149 PENFIELD, W Diencephalic autonomic epilepsy. Arch Neurol. & Psychiat, 22: 358, 1928
150 PINCOFFS, M. G. Case of paroxysmal hypertension associated with suprarenal tumor. Tr. Assoc. Am Phys., 44: 295, 1929
151 PITCAIRN, D M. AND YOUMANS, W. B Potent pressor action of an extract of a phaeochromocytoma after adrenolytic doses of dibenamine Fed. Proc, 8: 127, 1949
152 PITCAIRN, D M AND YOUMANS, W B The nature of pressor substances in pheochromocytomas Circulation, 2: 505, 1950
153 PLACE, V A. Piperoxan hydrochloride (benzodioxan) test report of a false positive reaction. J A M A, 146: 1227, 1951
154 PRATT, W A Chromaffin cell tumor simulating malignant hypertension J. Clin Endocrinol, 11: 630, 1951.
155 PRIESTLEY, J. T., SPRAGUE, R G, WATERS, W AND SALASSA, R M Subtotal adrenalectomy for Cushing's syndrome Ann Surg, 134: 464, 1951
156. RAAB, W Adreno-sympathogenic heart disease (neurohormonal factors in pathogenesis and treatment) Ann Int Med, 28: 1010, 1948
157 RABIN, C B Chromaffin tumor of the suprarenal medulla (pheochromocytoma) Arch Pathol, 7: 228, 1929
158 RANGES, H A AND BRADLEY, S E Systemic and renal circulatory changes following the administration of adrenin, ephedrine, and paredrinol in normal man J Clin Invest, 22: 687, 1943
159 REID, T M AND SALM, R A case of bilateral phaeochromocytoma Brit Med J, 1: 1116, 1949
160 RICHARDS, V AND HATCH, F N Surgical experiences with pheochromocytoma Ann Surg, 134: 40, 1951
161 ROSENBLUETH, A The Transmission of Nerve Impulses at Neuroeffector Junctions and Peripheral Synapses Technology Press, M I T, and John Wiley and Sons, New York, 1950
162 ROSENBLUETH, A AND CANNON, W B Adequacy of chemical theory of smooth muscle excitation Am J Physiol, 116: 414, 1936
163 ROTH, G M AND KVALE, W F Tentative test for pheochromocytoma Am J Med Sci, 210: 653, 1945
164 ROTH, G M AND KVALE, W F Further studies with histamine test for pheochromocytoma *Proc Central Soc Clin Res* 20: 47, 1947
165 ROTH, G M AND KVALE, W F Pharmacologic tests as an aid in diagnosis of pheochromocytoma Mod Concepts Cardiovas Dis, 18: 41, 1949
166 SEED, J C AND McKAY, E A Inhibition of piperidinomethyl-3-benzodioxane (933F) of epinephrine vasopressor blockade produced by dibenzyl β-chlorethylamine Proc Soc Exp Biol & Med, 70: 724, 1949
167 SHAPIRO, A P., BAKER, H M, HOFFMAN, M S AND FERRIS, E B Pharmacologic and physiologic studies of a case of pheochromocytoma Am J Med, 10: 115, 1951
168 SHIPLEY, A M Paroxysmal hypertension associated with tumor of suprarenal Ann Surg, 90: 742, 1929

Respiratory Failure in Neuromuscular Disorders

FRED PLUM, M.D.

In man, survival from extensive neuromuscular disease usually depends on the maintenance of homeostasis of oxygen and carbon dioxide. Death from poliomyelitis, the toxic and infectious polyneuritides, drug-refractory myasthenia gravis, or narcotic and anesthetic drug poisoning usually results from respiratory failure, and is frequently preventable if prophylactic and corrective treatment is efficiently and appropriately administered.

The development of a mechanical respirator by P. Drinker and Shaw in 1929 represented probably the greatest single contribution toward maintaining life in patients with paralytic respiratory insufficiency. Since the time this respirator was introduced, clinical and laboratory studies have widened our knowledge and understanding of the complexities of pulmonary physiology and respiratory gas exchange. Many of the results of these studies have been applicable to the challenging problem of artificial respiration and have resulted in better care and fewer complications in respirator patients. More recently a large number of mechanical devices have been introduced for artificial respiration, since less cumbersome apparatus than the "iron lung" has been desirable The ability to apply many of the newer physiological concepts and to assess the reliability of these new pieces of equipment has been necessarily limited to those institutions caring for large numbers of patients with paralytic respiratory insufficiency At the New York Hospital-Cornell Medical Center clinical observations on such patients have been augmented by and correlated with pulmonary function studies and metabolic and blood chemical analyses Physiological studies on human volunteers and experimental animals have supplemented this work, and the significant experiences reported by others dealing with the problem have been reviewed In the Cornell series, as with most others, acute poliomyelitis was the illness most commonly causing paralytic respiratory insufficiency. However, acute polyneuritis, myasthenia gravis, and poisoning by drugs which depressed neural function or conduction (morphine, barbiturates, curare), created a significant though smaller number of respiratory emergencies Over 75 per cent of all these patients survived

Neuromuscular respiratory failure also resulted from intracranial neoplasms, cerebral vascular accidents, or head trauma Apnea which resulted from structural displacement or extensive tissue necrosis of the brain was, however, seldom reversible, and treatment was usually fruitless in this group.

In: Methods in Medical Research, 3: 131, Year Book Publishers, Chicago, 1950.
190. VON EULER, U. S. AND HAMBERG, V. Colorimetric determination of noradrenaline and adrenaline. Acta physiol. Scand., 19: 74, 1949.
191. VON NEUSSER, E. Die Erkrankungen der Nebennieren, ed. 2, p. 97, Alfred Hölder, Vienna, 1910
192. WAALER, E Chromaffin tumor simulating toxic diffuse goiter Acta Med Scand, 123: 1, 1945
193 WADA, M Sudorific action of adrenalin on the human sweat glands and determination of their excitability. Science, 111: 376, 1950
194 WASHINGTON, E. L., CALLAHAN, W. P., JR AND EDWARDS, E. W. Pheochromocytoma of the adrenal medulla its role in the pathogenesis of a malignant hypertension J Clin. Endocrinol, 6: 688, 1946.
195 WATERS, L L AND DESUTO-NAGY, G. I Lesions of the coronary arteries and great vessels of the dog following injection of adrenalin Their prevention by dibenamine Science, 111: 634, 1950
196. WEGRIA, R Pharmacology of the coronary circulation Pharmacol. Rev, 3: 197, 1951.
197 WELLS, A H AND BOWMAN, P. G Clinical and pathologic identity of pheochromocytoma report of a case J A M A, 109: 1176, 1937.
198 WEST, G B Quantitative studies of adrenaline and noradrenaline J Physiol, 106: 418, 1947.
199 WILKINS, R W, GREER, W E R, CULBERTSON, J. W, HALPERIN, M. H., LITTER, J, BURNETT, C H AND SMITHWICK, R. H Extensive laboratory studies of a patient with pheochromocytoma before and after successful operation With a note on the trial of piperidyl-methyl benzodioxane to differentiate such conditions from essential hypertensive vascular disease Arch Int. Med, 86: 51, 1950
200. WILSON, G M Note on negative benzodioxane test. Lancet, 258: 761, 1950.
201 WUNSCH, R E, WARNKE, R D AND MYERS, G B The effects of dibenamine on severe hypertension Ann Int Med, 33: 613, 1950

whether caused by an infection, a metabolic illness or a toxic drug, is the result of a defect in one or both of these functions.

VENTILATORY FAILURE

Ventilation is the first respiratory function to be impaired in neuromuscular disease, and becomes inadequate (fig. 1) when interruption of innervation or control of the muscles responsible for breathing takes place,

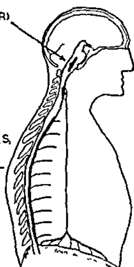


FIG. 1. VENTILATORY FAILURE IN NEUROMUSCULAR ILLNESSES

or when the muscles themselves fail As noted by Elam and his co-workers (18), this loss of innervation may be *peripheral* due to anterior horn cell degeneration, peripheral nerve disease, or defective neuromuscular transmission, or it may be *central*, the result of dysfunction or depression of the medullary respiratory center Either peripheral or central disorders result in progressively less air being moved in and out of the chest per unit of time, and therefore reduce alveolar ventilation.

In *peripheral failure* vital capacity falls as the muscles responsible for breathing progressively lose their innervation As the vital capacity falls, the breathing reserve is progressively impaired and tidal volume becomes more shallow as well The respiratory rate increases in an attempt to maintain respiratory compensation Sooner or later, dyspnea occurs as the demand for oxygen exceeds the patient's enfeebled breathing capacity so

All these disorders of the neuromuscular system produced respiratory insufficiency by essentially similar mechanisms which could be analyzed readily in terms of present concepts of respiratory physiology. Treatment in each case was more effective if guided by these principles. Variations from illness to illness were of secondary importance in treating respiratory failure. It rapidly became apparent that when any indication for active treatment existed, whether substitution of a respirator for ineffective ventilation or elimination of respiratory obstruction by tracheo-bronchial intubation, temporizing added to the danger to the patient. An accurate grasp of the pathological physiological alterations and the *immediate* institution of whole-hearted measures to correct them offered the greatest protection to the patient's life. Artificial respiration properly used in itself did not expose the patient to great hazard.

This monograph is divided into two main parts The first section is devoted to a review of theoretical and experimental data necessary for a clear understanding of the development and treatment of respiratory insufficiency This part considers physiological mechanisms of respiratory failure, methods of observation, and the principles and mechanical devices for producing artificial respiration. The second section is a step by step outline of clinical management which attempts to anticipate conditions arising at the bedside and describes the most effective procedures for dealing with them. The aim is to prevent decompensation of respiratory gas exchange in disease states affecting the neuromuscular system Methods of correcting anoxemia and hypercapnia are included, but with emphasis on prevention

The chief purpose is to clarify the mechanisms involved in neuromuscular respiratory insufficiency and to offer a guide to straightforward, practical and dependable treatment At present a very low mortality rate has been achieved in some institutions The same salutary results can be achieved in communities where physicians have had less of an opportunity to study these illnesses personally, but who at any time may be called upon to treat a patient with one of them.

MECHANISMS, DETECTION, AND METHODS OF CORRECTION

Mechanisms of Respiratory Failure

Baldwin, Cournand, and Richards (2) succinctly classified pulmonary function, into *ventilatory* function, the product of the chest bellows mechanism, which is responsible for the mass movement of air in and out of the lungs and *alveolar-respiratory* function which is concerned with the dis- [illegible] between [illegible] lure.

ness, saliva accumulates in the pharynx and is inhaled into the lungs Also, either as the result of failing ventilation or severe depression of consciousness (as in barbiturate poisoning), coughing becomes impossible so that not only is inhaled material not ejected from the lungs, but also the normal output of the tracheobronchial mucous membranes cannot be effectively raised and collects in the tracheobronchial tree. As these secretions accumulate they act as irritants, and stimulate the production of more mucus, intensifying the process. Secondary bacterial infection and exudation commonly follow. The customary attempts to clear saliva from the pharynx by external suction is irritating and increases salivary and mucus membrane outflow. Finally, if hypoxia occurs, capillary permeability is increased, and

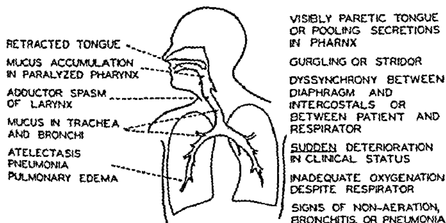


FIG 2 ALVEOLAR-RESPIRATORY FAILURE IN NEUROMUSCULAR ILLNESSES

still more mucus pours out Many of these phenomena are unavoidable in patients with paralyzed breathing and impaired swallowing mechanisms If they are compounded with other causes of obstruction such as a preexisting respiratory infection, a paralyzed lagging tongue, vomiting, or total pharyngeal or laryngeal immobility, the danger of blocking the airway is imminent If such block is not prevented by active measures, pneumonitis, atelectasis or pulmonary edema ensues The physiological distribution of the respiratory gases in the lungs is then impaired, and diffusion of carbon dioxide and oxygen between the alveolar capillaries and alveoli is prevented Alveolar-respiratory failure causes hypercapnia and anoxemia.

The Effects of Respiratory Failure

In humans, the arterial and consequently the tissue concentrations of respiratory gases are maintained normally between narrow limits Marked

that ultimately carbon dioxide in the blood is increased while the concentration of oxygen is reduced.

In *central* respiratory failure (dysfunction or depression of the brain stem respiratory center), uncomplicated by disease of segmental neuromuscular units, irregularities of respiratory rate and depth develop There is often difficulty in maintaining synchronous contraction of the diaphragm and intercostal muscles. Progressive slowing of the respiratory rate ensues and intermittent periods of apnea appear and become longer and longer. Occasionally, as Sarnoff, Whittenberger and Affeldt (55) have noted, the sensitivity to carbon dioxide as a respiratory stimulant is lost, though the ability to breathe deeply when commanded to do so is retained

If defects in both peripheral and central ventilatory function are present, centrally produced respiratory irregularities will be superimposed on shallow breathing and a low vital capacity. When irregularity of the rhythm or slowing of the respiratory rate become so pronounced as to reduce alveolar ventilation, hypoxia and hypercapnia result.

ALVEOLAR-RESPIRATORY FAILURE

Alveolar-respiratory failure (defective distribution of the respiratory gases in the lungs and impaired diffusion to the alveolar capillaries) is present in varying degree in any patient with advanced paralytic respiratory insufficiency (fig 2), usually being superimposed on ventilatory failure.

The most significant cause of disturbed alveolar-respiratory function in patients with paralytic respiratory failure is the accumulation of normally protective mucus secretions to a degree where they obstruct or threaten to obstruct the airway *

The excessive mucus secretions result from loss of the normal mechanisms for mucus removal rather than unusually large production. When swallowing is impaired, by pharyngeal paralysis or loss of conscious-

* Actually partial obstruction of the upper trachea or large bronchi, if not associated with excess secretions, may have as its principal effect reduction in tidal volume, and therefore may interfere primarily with ventilatory function Since such uncomplicated obstruction seldom occurs in the illnesses under consideration, all forms of obstruction are considered as interfering principally with alveolar-respiratory function

While it is recognized that in intrinsic pulmonary disease other pathological [illegible]

[illegible]

group which could not be explained on the basis of paralyzed expiratory breathing At the time of their studies, none of their patients had any airway obstruction

THE DEVELOPMENT, EFFECTS AND RECOGNITION OF HYPERCAPNIA

Although the dangers of hypoxia are considerable, the condition is a familiar one and most physicians are alert to it. In contrast, the possibility of carbon dioxide retention in the absence of anoxia is rarely thought of but nevertheless can cause dangerous depression of bodily functions, as shown by Bower et al. (6) and in our own studies. Hypercapnia is an insidious and constant threat to patients with ventilatory failure.

The mechanism of carbon dioxide accumulation in these patients is diagrammed in figure 3. As the tidal volume falls from paralysis of the respiratory musculature, respiratory rate increases in an attempt to sustain effective ventilation If, as in the patient hypothetically described in figure 3, respiratory rate is tripled to compensate for a two-thirds reduction in tidal volume, the minute volume may remain the same but the amount of the minute volume which merely flushes the respiratory dead space and does not participate in respiratory gas exchange is also tripled. Much of the effort of breathing is wasted in filling dead space. Eventually, tidal volume falls so low that the respiratory rate can no longer be increased enough to compensate. As a result, alveolar ventilation, the only part of the minute volume which exchanges oxygen and carbon dioxide with the blood, is reduced and alveolar partial pressure of oxygen falls and alveolar partial pressure of carbon dioxide rises With lowered alveolar ventilation, anoxemia is delayed or prevented by the affinity of hemoglobin for oxygen at lowered oxygen tensions. This physiological protective mechanism is usually enhanced by oxygen therapy which raises the partial pressure of oxygen in the inspired air.

Though anoxemia may thus be prevented no such protective mechanisms operate against the accumulation of carbon dioxide in the patient who is unable to increase his ventilation spontaneously Oxygen, a lighter gas, diffuses into the alveoli more readily than CO_2 diffuses out As carbon dioxide levels increase in the blood over a period of hours even the protective symptom of dyspnea may disappear, since prolonged exposure to elevated concentrations of carbon dioxide eventually reduces the sensitivity of the respiratory center and may actually depress the center.

Carbon dioxide in increasing concentrations acts first as a narcotic and then as an anesthetic (32, 58). Mental obtundation, disorientation, delirium and stupor have been reversed in some of our own patients with paralytic respiratory insufficiency when it was realized that these signs were due to carbon dioxide narcosis and not "polioencephalitis". This improvement followed increased ventilation and paralleled reductions in elevated blood carbon dioxide contents Insufficient pulmonary elimination of carbon dioxide results in a shift of blood pH to more acid levels since the gas is

deviations beyond these limits, particularly if rapid in development, cause pronounced changes in biologic function which may be fatal. In addition, relatively small degrees of anoxemia (lowered arterial oxygenation) or hypercapnia (excessive blood carbon dioxide) have the effect of producing tissue changes which directly interfere with the normal efficiency of pulmonary gas exchange and aggravate respiratory decompensation.

The effects of anoxemia and hypercapnia may be difficult to separate in observation at the bedside, but the two states can be delineated in certain clinical conditions as well as under experimentally controlled environments. In high altitude studies, for example, it has been possible to observe the effects of anoxemia without increased concentrations of carbon dioxide. Conversely in chronic pulmonary emphysema, during anesthesia and under controlled conditions of administering high concentrations of carbon dioxide, the effects of hypercapnia without anoxemia have been noted.

THE EFFECTS AND RECOGNITION OF ANOXEMIA

The earliest manifestation of anoxemia is increased capillary permeability (29) C. K Drinker (16) demonstrated that this response in the lungs is an accompaniment of relatively slight degrees of acute anoxemia and leads to pulmonary edema which further impairs respiration and aggravates the hypoxic state As widespread capillary damage takes place, circulation is interfered with, the pulse rate climbs, and eventually cardiovascular collapse and shock occur. The supervention of shock in acute respiratory failure is a dangerous complication and usually an irreversible one.

The clinical manifestations of hypoxia are subtle if the degree of hypoxia is slight Cyanosis, which has been used as the classical sign of oxygen lack, depends on the presence of at least 5 grams of unsaturated hemoglobin per 100 cc. of blood (35). Cyanosis, since it cannot be detected with any reliability with arterial oxyhemoglobin saturations above 80 per cent (11), is evidence of advanced respiratory decompensation and a sign to be avoided rather than to be used to guide treatment

Manifestations of less advanced impairment of oxygenation are an increasing pulse rate and apprehension, euphoria or emotional instability An increase in nasal, pharyngeal and tracheobronchial secretions is frequent in mildly hypoxic patients. The mildly hypoxic patient's color, though not assuming a cyanotic hue, blanches and appears grayer than when he is well oxygenated. Superficial cutaneous circulation appears depressed, perhaps because blood is directed to biologically more important organs. Any or all of these manifestations call for the assumption that oxygen intake is insufficient.

Carbon dioxide thus accumulates in toxic concentrations in patients with paralytic respiratory insufficiency, and has physiological effects which can be lethal. Hypercapnia may occur without anoxemia, especially during oxygen therapy, and should be anticipated in any patient with acute paralytic respiratory insufficiency. Correction of carbon dioxide retention in these patients can be achieved only by increasing pulmonary ventilation

Carbon dioxide retention may be suspected as the cause of dulling of consciousness or of hypertension in any patient with failing neuromuscular control of breathing However, sleepiness or an elevated blood pressure may be manifestations of the primary nervous system illness (37) so that the clinical impression of carbon dioxide retention must be confirmed by chemical analysis. The most accurate method of determining failure of CO_2 excretion is by direct measurement, determining the pH and CO_2 content or the partial pressure of carbon dioxide in the arterial blood. The cruder method of measuring the venous plasma CO_2 combining power along with a determination of urinary pH is a fairly reliable guide to CO_2 exchange. An increasing venous plasma CO_2 combining power, largely a measure of bicarbonate, reflects a compensation for respiratory acidosis and closely follows the development of hypercapnia. The high urinary chloride effected in attempting this compensation results in strongly acid pH of the urine

Detection of Respiratory Failure

Since respiratory insufficiency in neuromuscular illness may result either from failing ventilatory function or from interference with distribution and diffusion of the respiratory gases in the lungs, both functions must be adequate to prevent hypoxia and hypercapnia

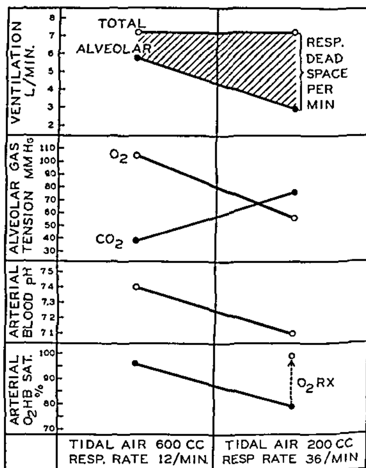
VENTILATORY FAILURE

An appraisal of peripheral ventilatory function should be made in all patients with acute paralytic illnesses by measuring vital capacity and tidal volume as early as possible and repeatedly at regular (8–12 hour) intervals until the acute stage of the illness has passed

Vital capacity and tidal volume recordings are easily made with a calibrated spirometer of the type used for BMR determinations Observations of this nature can be made as part of doctors' regular morning and evening rounds If a BMR instrument is not available, any reasonably accurate spirometer or vital capacity bellows will provide the necessary information. Special care must be taken to instruct the patient in the technique and importance of the procedure, as wide variations result if cooperation is not insured For children it may be helpful to make a game of the procedure in order to secure a true vital capacity.

Tables containing expected values for tidal volume and vital capacity

present in blood as carbonic acid, creating the condition termed respiratory acidosis. Although moderate increases in the arterial carbon dioxide concentration are associated with hypertension (22, 4) more marked hyper-



capnia produces cardiovascular collapse (4, 47) Recently, McDowell and Lee, working in our own laboratories, have demonstrated a significant amount of vaso-excitor-material (VEM of Shorr et al) paralleling the development of hypercapnia and hypertension in a patient with marked respiratory insufficiency This elaboration of a humoral substance in hypercapnia may explain, in part, the cardiovascular influence of this state

causes of breathing difficulties had their origin at different levels of the nervous system the results of ventilatory failure were the same: both developed CO_2 retention, and both recovered following relatively brief periods of artificial respiration.

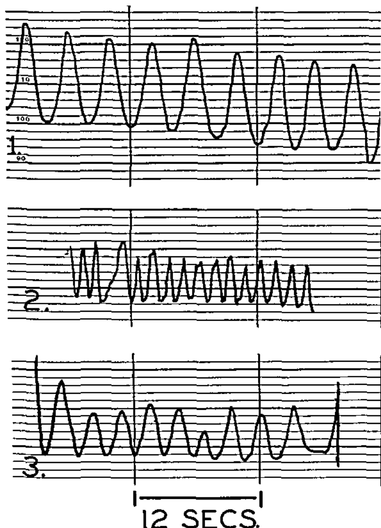


FIG. [illegible] [illegible] PATIENTS WITH [illegible]

(1) [illegible] n a patient with [illegible]

respiratory stimulant

exist for most age groups and may be used for reference normals (33, 52, 71). By making repeated observations it is possible to predict ventilatory failure many hours before it becomes apparent clinically so that proper precautionary and therapeutic measures anticipate an emergency. Bedside estimates of ventilatory exchange *without objective measurement* are grossly inaccurate and cannot be depended upon for such important information

It has been found by Plum and Whedon (1951) that as long as vital capacity stays above 1200 cc. in adults no significant impairment of respiratory gas exchange occurs, while if the vital capacity falls to 1000 cc or less during the acute stage of poliomyelitis or acute polyneuritis, artificial respiration for at least a few hours is nearly always necessary.

If a spirometer is unavailable one must depend on less accurate signs of failing ventilation such as an increasing respiratory rate, which usually appears before a visible reduction of the chest's excursion in breathing. As failure progresses, patients become reluctant to talk, and coughing is at first weakened and then virtually ineffective. In adults, when forceful coughing is lost, due to loss of expiratory volume, the vital capacity seldom measures more than 1200 cc.

Another clinical test which provides a little comparative information is to ask the patient to count progressively on one full expiration. Healthy adults counting slowly should reach at least thirty By having patients repeat this maneuver at regular intervals and comparing one performance with the next, the approximate reduction in ventilation can be ascertained. For example, if a patient had been able to count to thirty-four on admission, and later was able to count to only seventeen, it would be assumed that the vital capacity was reduced by about half.

On the basis of studies correlating these clinical signs with spirometric readings in our patients, we found that artificial ventilation is generally necessary if the respiratory rate climbs above thirty-five, if the expulsive force of coughing has practically disappeared, or if the patient can count less than ten.

In contrast to the manifestations of peripheral ventilatory failure, failure of *central* ventilatory control is manifested by a progressively slower rather than faster respiratory rate The diaphragms and intercostal muscles frequently fail to contract synchronously and there occur irregularities in rate and depth of respiration. Such irregularities when severe may be apparent on inspection, but their accurate and early detection should depend on reliable spirometric tracings Figure 4 demonstrates a typical tracing from a patient with central respiratory failure (#3 of fig 4). Despite a good vital capacity, this boy had an insufficient minute alveolar volume to accomplish adequate CO_2 elimination Compare this tracing with the example from a patient with peripheral ventilatory failure (#2 of fig 4) Though the

not caused physical signs in the chest, bronchial obstruction may be suspected when a patient in respiratory distress suddenly displays an increase in pulse and temperature, or deteriorates clinically at a rate faster than accounted for by his paralysis. Sudden increase in dyspnea usually indicates obstruction of one of the larger radicals of the bronchial tree, although in rare instances abrupt failure of the respiratory center may be the cause. Also, if adequate ventilation or sufficient oxygenation and carbon dioxide elimination cannot be achieved by using maximum negative pressures of 20 to 25 cm. water in tank respirators with cycling rates set at 20 to 24 per minute, it can be safely concluded that the airway is not clear.

Treatment of Respiratory Failure

When evidence of marked deterioration of ventilatory function or impairment of gas exchange by obstruction of the respiratory passageways develops, corrective treatment must be immediately instituted. Failing ventilation must be supplemented or replaced by some type of respirator; and obstruction must be removed either by suctioning out the pharynx or, if necessary, by bronchoscopy and tracheotomy Oxygen therapy, which is sometimes used in an attempt to delay the need for a respirator or to avoid a tracheotomy, is of doubtful value at this stage. Oxygen, by preventing anoxemia in a state of under-ventilation, permits unsuspected and dangerous hypercapnia to progress Since oxygen is usually given by nasal catheter, it often irritates the mucous membranes of the pharynx, increases the amount of secretions and adds to the likelihood of obstruction developing In most instances, as Moyer and Beecher (43) have found, if ventilation *is so markedly reduced that oxygen is necessary to prevent anoxemia*, inadequate removal of carbon dioxide may already exist This will be true particularly in barbiturate poisoning, which is chiefly a problem in neuromuscular respiratory depression (45), for barbiturates selectively depress the respiratory center's response to carbon dioxide For these reasons, if oxygen therapy is needed to prevent anoxemia in neuromuscular ventilatory failure, artificial respiration should be started at the same time to insure carbon dioxide elimination. Similarly, if obstruction of the airway has progressed to a stage where oxygen is necessary, the obstruction should be eliminated by tracheotomy, before oxygen is relied upon

TREATMENT OF VENTILATORY FAILURE

The treatment of a failing ventilation is the substitution of extrinsic manual or mechanical control for the failing intrinsic control of the pa-

tion depends on

iere and alveoli

Cyclic pressure

ALVEOLAR-RESPIRATORY FAILURE

Interference with alveolar-respiratory function in acute neuromuscular illnesses is nearly always due to obstruction of the respiratory passageways Since no single clinical or laboratory test will detect such obstruction, impairment of alveolar-respiratory function is considerably more difficult to detect than impairment of ventilatory function, and is easily overlooked if not constantly sought for To a certain degree, the manifestations of partial or complete blocking of the respiratory passageways vary with the site which is threatened Puddling secretions in the nasopharynx usually are visible if the pharynx is inspected and generally are so annoying to the patient that he complains. Paralysis of the tongue is similarly evident. The gradual accumulation of mucus at deeper levels of the respiratory passageways advertises itself less loudly, however, and produces symptoms and signs which are often quite subtle or mimic the changes seen with actual neural defects.

Unfortunately gurgling respirations, stridor or audible laryngeal and tracheal rattles are uncommon even with large collections of fluid in the hypopharynx, the laryngeal crypts, or the trachea, so that trouble is often not suspected until obstruction is nearly complete Puddling at these levels should be suspected in any patient with pharyngeal paralysis. In addition most patients who have progressed to a stage of respiratory insufficiency or who have lost the ability to cough within less than 24 to 36 hours of the onset of paralysis have excessive unremoved mucus although they may have no weakness of swallowing Patients with such difficulties protect themselves, often subconsciously, against inspiration of material deeper into their lungs They breathe shallowly, so as to pass the stream of air over the fluid, and often breathe irregularly in depth and rate for the same reason. The normal synchrony of diaphragmatic and intercostal muscular action is often lost presumably because of the care with which they must breathe. Since these symptoms of respiratory irregularity and the loss of synchrony from partial airway blockage are identical with breathing patterns which result from central ventilatory failure, correct differential evaluation must be based on direct inspection of the upper airway through a laryngoscope Laryngoscopy is a relatively simple and non-traumatic procedure It may be repeated every few hours without excessively distressing the patient Bronchoscopy is not recommended as a diagnostic procedure unless it is to be followed immediately by tracheotomy since it is followed by a marked increase in secretions of the mucus membranes.

Partial or complete encroachment on the bronchial lumen deep in the lungs usually cannot be identified until it becomes extensive and results in atelectasis and pneumonitis If pulmonary consolidation or infection has

the Holger-Neilsen (1932) method for manual artificial respiration. Although the Neilsen method did not produce as much ventilation as alternate raising of the hips and compression of the back, it required less energy, was easier to teach, and was less variable during application by a rescue worker. For all these reasons the Neilsen method has been adopted.

The following description of the technic is an excerpt from the report of the Council on Physical Medicine and Rehabilitation of the American Medical Association (14): "The subject is placed prone, elbows bent, arms overhead with one hand upon the other. The cheek is placed upon the hand, the face turned slightly to one side. The operator kneels on one knee at the head of the subject and puts the foot of the opposite leg near the (patient's) elbow. He places his hands on the subject's back in such a way that the thumbs just touch, the heels of the hands being just below a line running between the armpits. He (the operator) rocks slowly forward, elbows straight, until his arms are approximately vertical, exerting steady pressure upon the chest Then he commences to rock backward slowly and slides his hands to the subject's arms just below the elbows He raises the arms until resistance and tension are felt at the subject's shoulders. Then he drops the arms. This completes a full cycle. The cycles are repeated 12 times per minute, the expansion and compression phase being of equal length, the release periods being of minimum duration A standard technique for administering the method is being prepared by a committee representing many organizations."

Nims, Comroe and co-workers (46) feel that a respiratory rate of 12 times a minute as recommended may be insufficient to ventilate asphyxiated patients adequately using the Neilsen method (or any manual technic) It is their opinion that, since underventilation is so dangerous, a more rapid rate should be employed.

Rocking Method of Artificial Ventilation

Alternately tilting the prone or supine patient from the head-down to the foot-down position and back has been widely used as a method of artificial respiration since Eve in 1932 used a rocking chair to resuscitate a child with ventilatory failure caused by diphtheritic polyneuritis Comroe and Dripps (12) and Asmussen and Neilsen (1), have shown that rocking produces tidal volumes that increase in direct proportion to the amount of arc through which the patient is tilted. For emergency artificial respiration, rocking arcs of a minimum of 45 to 60 degrees each way from the horizontal (90 to 120 degrees total arc) must be used, lesser arcs providing insufficient ventilation in patients who are unable to supplement the action of the rocker by intrinsic ventilatory efforts Probably greater ventilation can be produced by this method than by any of the purely manual techniques,

gradients are usually established by (a) intermittently raising the air pressure at the oral end of the airway (intermittent positive pressure breathing, IPPB), (b) alternately raising and lowering the air pressure of the oral airway ("suck and blow" respirators), or (c) alternately decreasing and increasing the external pressure about the chest and/or diaphragm with the upper airway exposed to the atmosphere (for example, by tank respirator). Evidence has been advanced by Maloney and Whittenberger (36) that all these methods achieve essentially equal ventilation and the same physiological effects if equivalent pressures and pressure curves are used. This point has been disputed, but each method can certainly produce adequate ventilation when used properly.

Several methods of producing active artificial inspiration by intermittently decreasing the external pressure about the chest and diaphragm have been tried on our patients, sometimes combined with methods of actively achieving expiration. Manual artificial respiration, the rapid rocking bed, tank type (iron lung) respirators, cuirass respirators and the electrophrenic respirator have all been evaluated. When sufficient data were available the absolute and comparative efficiency of each of these methods were assessed Since a great deal of work has been done recently by others interested in these methods, the results of many studies have been incorporated in the following sections which describe these various devices and technics.

Manual Artificial Respiration

This becomes necessary for patients with acute paralytic respiratory insufficiency when mechanical equipment is unavailable or fails

Pioneer experiments (10, 12, 13, 24, 63) indicated that the time-honored Schafer (1909) method of artificial respiration produces ventilation inadequate for asphyxiated patients The potential danger, in case of war, of large numbers of respiratory casualties due to nerve gas has led to extensive investigation and reevaluation of methods of manual artificial respiration. As a result of these studies, originated largely by Gordon and his coworkers (23, 24), and supplemented by groups at Harvard University, University of Pennsylvania and Springfield College, it has become apparent that artificial respiration which produces both active expiration and inspiration is two to three times as effective as one which produces either alone Since the Schafer method produces only active expiration, and depends for inspiration on elastic thoracic and pulmonary recoil (which may be negligible in asphyxia), it is one of the least effective methods, and is inadequate for maintaining oxygen saturations even in normal individuals rendered temporarily apneic by curare (25)

Therefore, the American Red Cross, The Department of Defense, and the American Medical Association have replaced the Schafer method with

leaving much space between the plastic dome and the patient, their edges fit snugly to the patient with adjustable rubber edges. The cuirasses are comparatively small and uncumbersome and are supplied with intermittent negative and/or positive pressure from a separate pump, accomplishing breathing by creating negative or positive pressure about the patient's chest and abdomen. As originally noted by Plum and Lukas (48) cuirasses produce considerably (20 to 50 per cent) lower tidal volumes than do tank respirators operated at equivalent pressures. They have thus proved inadequate for ventilating many patients throughout their acute illnesses. This comparatively lower ventilation apparently is due to the fact that few cuirasses induce any lateral thoracic or abdominal breathing movements Also, since they tend to constrict upper and lateral thoracic movement they are not recommended for continuous use at any time, lest muscle contractures result.

If these limitations of the ciurass respirator are heeded, they have many advantages. They do produce enough ventilation in nearly all patients acutely ill with paralytic respiratory insufficiency to supply artificial respiration during short periods of emergency transportation. They will also ventilate most acutely ill patients sufficiently to allow them to be removed from a tank respirator for a tracheotomy. Since patients convalescing from acute paralytic respiratory insufficiency usually have lowered respiratory demands and more flexible thoracic and abdominal walls, cuirass respirators will provide long periods of adequate ventilation for nearly all of this group and permit a degree of freedom of activity unattainable in tank respirators. In this way, the cuirass accelerates convalescence and makes it easier. Whenever a measured safe ventilation can be achieved, cuirass respirators are of assistance in managing patients with paralytic respiratory insufficiency. Since they provide less ventilation than tank respirators, some discrimination is necessary in their application.

In using cuirasses, higher pressures and faster rates are necessary than in tank respirators. The skin of the patient must be kept scrupulously dry where the cuirass is applied lest friction necrosis occur Talcum powder helps in this prophylaxis Persons with particularly sensitive skin may need soft flannel between them and the chest piece to prevent abrasion. Also leaks between the edge of the respirator and the patient's body can be sealed in this fashion.

Tank ("Iron Lung") Respirators

These devices still are the most reliable and safest for artificial respiration for most patients with ventilatory failure due to acute neuromuscular disease Recent efforts to develop other types of negative pressure respirators which are less cumbersome or embody some special features have re-

provided sufficient arc is used. Although Gordon's group (23, 24) found that rocking produced smaller ventilation than the hip-roll-prone pressure technic, they used such a small arc that their results were not comparable to those of other workers.

Mechanical rapid rocking beds are available for the care of patients with neuromuscular ventilatory failure and were first used for patients with poliomyelitis by Wright (72). Operating on the same principle as any manually driven tilt table, these devices are intended to provide artificial ventilation without incarcerating patients as in standard respirators. They have been designed to furnish comfortable respiratory assistance for several hours.

Studying the effects of these mechanical rapid rocking beds on patients with ventilatory insufficiency caused by poliomyelitis, Plum and Whedon (49) found (since they tilted through total arcs of only 40 to 44 degrees) they produced no more than 60 to 70 per cent of the amount of tidal air that could be achieved by a tank respirator Acutely ill, febrile patients retained carbon dioxide and became anoxemic while on a rocking bed and these defects had to be corrected by putting these patients in tank respirators. The rocking bed was therefore contraindicated for acutely ill patients. During convalescence, on the other hand, when metabolic demands were less, and patients could gradually adjust to the constant rocking movement, rapid rocking beds furnished excellent ventilation for all patients with ventilatory insufficiency, regardless of the degree of breathing paralysis. Adaptation to the rocking motion of the bed sometimes was difficult at first. Initial periods of rocking often had to be limited to a few minutes, but within two weeks nearly all patients were able to tolerate rocking comfortably for periods of one to four hours Periods of longer than four hours were possible in patients who had some return of their own powers of ventilation, fatigue and respiratory decompensation gradually developed in the extremely paralyzed Maximal rocking arcs (40 to 45 degrees) were necessary to provide ventilation. The rocking rate at first was adjusted to meet the subjective breathing demands of the patient, but as soon as possible tidal volume was measured with the spirometer to assure ventilation sufficient to prevent the gradual development of anoxemia or hypercapnia. Rocking rates of 16 to 25 per minute were necessary, depending on the degree to which patients were able to supplement the ventilatory effect of the bed The faster rates were required if chest fixation had taken place or if the paralysis of breathing musculature was extensive.

Cuirass Model Respirators

These respirators are plastic pieces (the cuirass) which fit over the anterior chest and at least the upper part of the anterior abdomen. Though

respirator cycle (for example, −8 cm H_2O, +8 cm. H_2O). However, in preliminary unpublished studies on dogs, Lukas and Plum have found that, although increasing amounts of negative respirator pressure progressively reduce the cardiac output, the subsequent addition of increments of intra-tank positive pressure to the respirator cycle does not significantly reverse the trend. Until more is known about this important matter it seems best to place greatest reliance on negative pressures which are more comfortable and which achieve greater ventilation. Moderate increments of intra-tank positive pressure may then be added within the limits of comfort to aid in expiration and perhaps reduce the likelihood of circulatory depression.

Electrophrenic Respirators

The electrophrenic respirator (EPR) introduced by Sarnoff and his colleagues (53–57) is still having its advantages and limitations investigated. This respirator which intermittently stimulates the diaphragm consists of a small control box which delivers graded electrical impulses through an electrode attached either to the cutaneous motor point of the phrenic nerve or directly to the nerve itself. Since diaphragmatic movement depends on the trophic integrity of the phrenic nerves and intact neuromuscular transmission, use of the electrophrenic respirator in the acute neuromuscular illnesses is limited to selected patients. However, the electrical stimuli applied to the phrenic nerve inhibit intrinsic phrenic stimulation For this reason, the patient with poliomyelitis who has involvement of the respiratory center without segmental degeneration of the anterior horn cells supplying the phrenic nerves may be markedly benefited. Similarly benefited will be other patients without segmental neural degeneration, such as those suffering from poisoning with anesthetic drugs. Circulatory depression in such patients appears to be reversed during EPR (53, 54), and the only hazard is that it is not possible to predict the time of degeneration of the phrenic nerve which may occur in any patient with poliomyelitis Little help from this device is likely for patients suffering from spinal poliomyelitis, from polyneuritis or from myasthenia gravis

The advantage of EPR as a method of artificial respiration for patients in whom the integrity of the phrenic nerve is assured is that the patient is left entirely free for nursing care Practically any desired amount of ventilation can be achieved by appropriately regulating the rate and intensity of electrical stimulation to the phrenic nerve Also, it is claimed by Sarnoff and his colleagues (53, 54) that other types of artificial respiration operating by cycling positive or negative pressure produce circulatory depression which is avoided by EPR The cutaneous motor point of the phrenic nerve is said by the original investigators (55, 57) to be found rapidly in a majority of subjects

sulted in useful supplementary apparatus, but have not equalled or supplanted the tank respirator. We have found that tank respirators produce larger tidal volumes than any other available respirator in unselected patients with paralytic respiratory insufficiency. They are easy to operate and maintain, and are familiar enough so that little special training is required to assure their proper use whenever the need arises (70). A wide range of tidal volumes can be achieved by altering respirator pressures. They cause little personal discomfort to the patient. Though pulmonary emphysema, pulmonary edema and alveolar hemorrhages have been noted sometimes at autopsy in patients dying after receiving artificial respiration (26), pulmonary function studies by Lukas and Plum (34) demonstrated that these changes were not common in surviving patients, and artificial respiration of itself did not commonly cause damage to the lungs when administered by tank respirator.

Since no moving respirator parts come in contact with the patient except at the neck, there is little hazard of abrasion of the skin or fixation of the chest. An additional advantage is that if electrical power fails, manual operation is comparatively easy and produces just as large tidal volumes as when the machine is working on its own motor. The only significant drawbacks are that it is difficult to examine the patient and nursing care is harder to apply to a patient within the tank respirator Also active or passive movement of the body or extremities is quite limited.

The purpose of these respirators is to provide ventilation Since the amount of respirator pressure needed to produce effective ventilation varies considerably from patient to patient it is desirable, if not imperative, to measure a patient's tidal volume at regular intervals when in a tank respirator Setting the apparatus to produce a certain pressure reading is no guarantee that adequate artificial respiration will be supplied Pressure gauges on respirators are not always reliable, often reading either too high or too low. In addition, wide variations in tidal volume have been observed using identical respirator pressures, both on different patients early in their illnesses and on the same patient at different stages of his disease Determination of tidal volumes by spirometer and the consequent adjustment of respirator pressures and rates to produce physiological ventilation prevents unrecognized under- or over-ventilation and serious consequences

The circulatory effect of total pressures and pressure curves created by various respirators has been scrutinized recently With the type of respirator pressure curve developed by tank respirators, cardiac output is reduced and it has been thought that this may contribute to the hypertension so commonly seen in respirator patients. Maloney and Whittenberger (36) believe that these depressing circulatory effects may be overcome by using equal amounts of positive and negative pressure on each

mittent positive pressure breathing are available for resuscitation. The most widely known is the standard closed system anesthesia set which supplies passive inflation of the lungs as the operator manually compresses the bag The advantages and technics of this method recently have been reviewed by Watrous, Davis and Anderson (64). The other device used extensively for patients with neuromuscular respiratory failure is the positive pressure unit designed by Bennett to connect with the bellows shaft of a Drinker respirator and described by Bower, Bennett, Dillon and Axelrod (6) Other types of intermittent positive pressure breathing respirators and the physiological effects of the methods have been discussed by Motley and his co-workers (41, 42) and by Barach, Fenn, Ferris and Schmidt (3). Though the effects of many of these devices are comparable, they are seldom used in the illnesses under consideration.

Thus, the advantages of intermittent positive pressure breathing are that it provides a method of ventilating patients who must be removed from a respirator or for whom a respirator is not immediately available. Intermittent positive pressure breathing when added to the tank respirator also increases ventilation in the few patients who are underventilated by maximal pressures and rates in the tank.

Operated properly, intermittent positive pressure breathing has few deleterious effects. The chief danger is that the method, improperly used, produces circulatory depression by decreasing cardiac output The reduction of cardiac output follows elevation of mean intrathoracic pressures which impairs venous return to the heart This effect can be minimized or abolished (15) if the pressure curve rapidly reaches peak pressure, rapidly falls off to atmospheric pressure and allows time for expiration longer than that required for inspiration. According to anesthesiologists (64), pulmonary damage, either in the form of pneumothorax, pulmonary, or mediastinal emphysema is but a slight risk if maximal positive pressures of 15 to 20 mm Hg are used. Though acapnia occasionally is produced because of excessive intermittent positive pressure breathing, this is a relatively benign state when mild and is hardly likely to occur in acute neuromuscular illnesses where alveolar-respiratory defects invariably interfere with respiration.

TREATMENT OF ALVEOLAR-RESPIRATORY FAILURE (OBSTRUCTION)

Since respiratory obstruction in paralytic insufficiency is almost wholly the result of the accumulation of secretions for which the normal mechanisms of removal are deficient, treatment is a straightforward problem of removing these fluids and preventing their recurrence. How extensive a therapeutic measure will be required depends entirely on the level of the pharyngeal-tracheal-bronchial airway which is threatened.

Disadvantages in EPR have been noted by other workers polled (1951) by the Sanborn Company who manufacture the apparatus. Many were unable consistently to locate the cutaneous motor point of the phrenic nerve. Surgical exposure with direct application of the electrode to the phrenic nerve in the neck frequently has been necessary to provide effective diaphragmatic stimulation. Such a surgical procedure may be time-consuming so that artificial respiration by another method must be started first. Spread of electrical stimulation frequently causes pain in the neck. Also, stimulation of diaphragmatic movement at times abruptly fails. This catastrophe has been attributed to depolarization of the electrode Since it has been unpredictable, a standby respirator of another type is needed when EPR is applied.

More widespread experience with EPR is needed before its indications and contraindications are settled upon. It cannot be recommended for general use at this time. It represents the practical development of a method of artificial respiration that has engaged speculation for years, and may eventually prove to be the most important contribution for patients with depressed breathing on a central basis since Drinker introduced the first tank respirator

Positive Pressure Breathing

Intermittent positive pressure breathing (IPPB) is not used as commonly as the more conventional methods of artificial respiration in acute neuromuscular diseases because it is uncomfortable to patients when applied for long periods of time The method also demands the full attention of at least one person Since, however, IPPB can be depended upon for adequate ventilation, it is sometimes a valuable adjunct to other kinds of respirator therapy. Delivered by face mask or to an endo-tracheal tube, intermittent positive pressure breathing will supply excellent ventilation so that a patient can be removed safely from a respirator for a tracheotomy to be performed or for some other maneuver to be carried out The great advantage is that the patient is unencumbered by equipment except for a face mask Also, when applied to a mask or tracheotomy tube in synchronization with intermittent reduction of pressure around the patient by a tank respirator, IPPB increases the external air-alveolar air pressure gradient and thus improves ventilation In convalescence, it is sometimes possible to create larger tidal volumes by positive pressure inflation of the chest than can be achieved with any available respirator When used in this fashion to passively expand the chest, positive pressure may increase the expansibility of the thorax and reduce the thoracic resistance to spontaneous breathing efforts.

Many devices which operate partly or wholly on the principle of inter-

With ventilatory failure, serious incoordination between a patient and the cycling action of the respirator often will be relieved by tracheotomy. Such incoordination may result from the patient's voluntary efforts to avoid inhaling pharyngeal secretions. It may also result from panic, inability to relax, or it may be a manifestation of respiratory center disease. That the respiratory musculature is too weak to resist the action of the respirator is unimportant in such instances, for effective ventilation is prevented by the glottis closing against the stream of inspired air. A tracheotomy obviates these difficulties, since it provides an orifice below any point that the patient can close voluntarily.

Tracheobronchitis or pneumonitis in patients with ventilatory failure is an indication for tracheotomy so that the muco-purulent outpourings can be removed from the tracheobronchial tree. Without external removal such exudative material will collect rapidly and cause asphyxiation.

Tank type respirators have been repeatedly demonstrated to produce sufficient ventilation to exchange oxygen and carbon dioxide in acutely ill patients without intrapulmonary lesions such as pneumonia, atelectasis or pulmonary edema If anoxemia or hypercapnia develops in a respirator patient receiving maximal respirator pressures and rates, it must be inferred that part of the lungs are not being ventilated, possibly as the result of a bronchial occlusion. Although bronchoscopy may re-establish the continuity of the airway temporarily, it is a traumatic procedure which itself stimulates mucus production so that obstruction is likely to recur. Tracheotomy with repeated endobronchial suction will avoid these difficulties

Tracheotomy is also desirable in two conditions in which actual or potential airway obstruction may be strongly inferred, although there may be no signs of its presence: Coma with acute poliomyelitis, polyneuritis or myasthenia is usually a sequel of inadequate respiratory gas exchange. Although such patients at times have been considered to have "encephalitis", they often improve dramatically after tracheotomy is performed, (as noted by Baker and his co-workers in Minneapolis, 1947), so that the chest may be cleared of occluding secretions and ventilation increased. The other condition for which tracheotomy should be considered is rapidly progressing poliomyelitis or polyneuritis which produces ventilatory failure within 24–36 hours of the onset of paralysis. Since at least in poliomyelitis, paralysis usually progresses for 3 or 4 days, there is no way of predicting which structures are to be affected. If not protected many patients with respiratory insufficiency develop pharyngeal paralysis and some suffer suddenly fatal obstruction. It is safer to have a tracheotomy in place before these individuals are in respirators Once a patient has suffered serious anoxia because his airway was blocked, his chances of survival are considerably reduced. In addition the technical difficulties of performing tracheotomy are greatly increased when the patient is in a respirator. If

For patients with impaired swallowing not requiring artificial respiration, use of the prone position with the foot of the bed elevated 20 degrees will allow excessive pharyngeal secretions to drain spontaneously If intermittent pharyngeal suction is applied gently, these measures should be adequate for this relatively uncomplicated condition.

Such simple measures will not suffice when breathing mechanisms are impaired seriously for the head-down position crowds the diaphragm with viscera and interferes with ventilation. If respirator care is necessary it is next to impossible to get the acutely ill patient on his face and also achieve any kind of effective artificial respiration. The development of respiratory obstruction is thus most difficult to prevent when the need for artificial respiration is present or impending.

Like most of the complications of paralytic respiratory insufficiency, airway obstruction is easier to prevent than to treat. For this reason and because of the rapidity with which death may follow airway closure, it is logical to safeguard the airway. Tracheotomy and deep suction serve better than temporary endotracheal intubation in a condition likely to last for several days. Although many patients with paralytic respiratory insufficiency do not need this additional safeguard, a large proportion do Tracheotomy should be performed with uncompromising directness when present or potential interference with the airway exists. Affirmation of this viewpoint is almost universal (20, 21, 38–40, 50, 62) and has been at least partly responsible for bringing the mortality rate among respirator cases to as low as 15 per cent in some poliomyelitis epidemics.

Tracheotomy is called for in a patient suffering from one of the acute neuromuscular illnesses whenever the integrity of gas exchange between the alveoli and outside air is threatened by paralysis of tongue, pharynx or larynx, depressed cough reflexes, or excessive mucous membrane secretions which cannot be cleared by simple pharyngeal suction. The need for the procedure is considerably greater in actual or impending ventilatory failure

Tracheotomy is needed in the following conditions:

When ventilatory failure develops in the presence of impaired swallowing, fluids accumulate in the pharynx and often are drawn into the trachea. Usually it is impossible for the patient to cough so material drains into his chest If artificial respiration is needed, the cycling action of the respirator tends to draw saliva into the tracheobronchial tree from the point where it accumulates in the hypopharynx The presence of a tracheotomy in such patients directs the main inspiratory stream below the puddles in the hypopharynx, and in addition allows direct suction of the tracheobronchial tree.

If bilateral abductor paralysis of the larynx (a rare phenomenon) occurs, immediate tracheotomy will be necessary to pass the obstruction and permit ventilation.

Physical Observations

Once the diagnosis of one of the acute neuromuscular diseases has been made, charts are posted where clearly visible on the wall beside the patient's bed for the purpose of providing immediately available data on the course of the illness for at least the preceding 24 hours. They contain the following observations:

1) Pulse is recorded at least every 4 hours. Tachycardia is an indication of continued activity in polyneuritis (27), an illness in which the temperature frequently is *not* elevated. Also, sudden changes in pulse rate may indicate hypoxia, a developing respiratory obstruction, or cardiac arrhythmia—a manifestation of disease of the brain stem

2) Blood pressure is recorded every 4 hours while the patient is awake Although hypertension is a frequent concomitant of poliomyelitis and is seen as well in some cases of polyneuritis, rising blood pressures may indicate retention of carbon dioxide On the other hand, hypotension may indicate severe depression of brain stem function in acute drug poisoning and thus call for increased ventilation.

3) Respirations are recorded every 4 hours routinely and more frequently with rates above 20 to 22 per minute. Special note is made of irregularity in rate or depth which might herald central respiratory failure.

4) Temperature is recorded every 4 hours Although not an absolutely reliable indicator of cessation of activity of poliomyelitis (patients occasionally deteriorate significantly in the post-febrile state), return of the temperature to normal usually means the danger period is over in this disease. Sudden rises in temperature in patients with respiratory difficulties may indicate pneumonitis or atelectasis

5) Fluid intake is charted. Since many patients with acute neuromuscular illnesses are anorexic, and some have nausea and vomiting, fluids are given parenterally if peroral intake is inadequate. The beneficial effect of dehydration on edema associated with inflammation of the nervous system is doubtful, and liberal hydration is sought

6) Urine output is noted. Many patients with acute neuromuscular diseases are unable to void spontaneously at some time in their illness due to either loss of motor power or proprioceptive sensibilities Excessive bladder *distention with its painful consequences* is avoided if the time and amount of urine are noted, alerting the staff to the possibility of urinary retention In addition, since the neuromuscular illnesses frequently occur in hot weather, relative dehydration may occur and be reflected in a low urine output, even when adequate fluids are drunk

7) Vital capacity and tidal volume are recorded on admission and twice daily thereafter at morning and evening rounds on every patient. The frequency of measurements is increased at the first sign of decreasing ven-

tracheotomy is performed before artificial respiration has been started, the lowest number of pulmonary complications occur.

The performance of tracheotomy needs little comment, since surgeons skilled in nose and throat surgery live in nearly every community Paralytic respiratory insufficiency imposes only two modifications on the usual procedure: as large a tracheotomy tube as possible (#5–#6) should be used so as to achieve the widest attainable airway, and the incision should be made between the second and fourth tracheal rings so that the tube will not be encroached upon by the collar of the respirator.

Tracheotomy is not always without harmful effects, so it is not done routinely for all patients with paralytic respiratory insufficiency. Being an operation, it requires additional persons and equipment, and leaves some slight disfigurement on the neck after healing. More important, the tube is an irritant and guarantees increased tracheal secretions, perhaps for long after the emergency has passed Also, some oozing of blood from about the fresh stoma always occurs, and in very rare instances frank hemorrhage into the trachea develops from an improperly ligated vessel and causes asphyxia. Occasionally if the tracheotomy tube is inserted low in the neck, the action of the respirator draws air into subcutaneous tissue resulting in subcutaneous emphysema and, rarely, mediastinal emphysema or pneumothorax

Any of these possible complications are fortunately rare, and hardly balance the tremendous therapeutic value of the artificial airway in properly selected cases When a tracheotomy is performed skillfully and cared for properly complications are so infrequent as to be practically non-existent and the procedure has saved the lives of many who could not have survived otherwise

A CLINICAL REGIMEN OF MANAGEMENT

Patients with respiratory failure from the acute neuromuscular diseases are so ill and may develop potentially fatal complications so rapidly that round-the-clock supervision is necessary Neither an efficient diagnostic laboratory nor the best of mechanical equipment can substitute for close clinical observation, and a trained, alert, always available staff

To prevent or treat respiratory failure, its presence must be known. Bedside clinical observation is inaccurate in evaluating the amount of respiration of which a patient is capable and must be supplemented by more reliable data. Therefore, the first step in the management of paralytic respiratory failure is the collection of reliable observations which detail the patient's condition and progress The following program, developed on the Cornell-New York Hospital Neurological Service, has proved reliable both in assessing the clinical state and administering treatment

4) Serum sodium and potassium analysis is rarely needed, as in the occasional respirator patient who must subsist for long periods on parenteral fluids. Neuromuscular illnesses, particularly poliomyelitis, are accompanied by an increased potassium loss in the early weeks, attributable in part to loss of muscle tissue (66). These losses are not much greater than those noted in other diseases where eating is impaired. Severe depletion of potassium, however, has been reported by Lans and his co-workers (31) in respirator patients when added daily increments of potassium salts were not administered. Further prostration and muscular weakness was added to that of the primary illness. Sodium depletion is less likely to occur than potassium deficiency because of the widespread use of saline in parenteral fluid therapy. In hot weather, however, and with the frequent use of hot packs to relieve pain, hyponatremia may be encountered.

5) Any patient with brain stem disease or with severe respiratory insufficiency is typed and cross matched and compatible whole blood placed on call Shock may appear suddenly and even though usually refractory to treatment, immediate whole blood transfusion occasionally has reversed the process. Also, the blood hematocrit is checked at intervals of 4 to 7 days in the more severely ill patients Infection inhibits red blood cell formation and patients with a significant fall in hematocrit in the second week of illness may need transfusion.

General Principles of Management

Since complete neuromuscular examinations are useless therapeutically and unreliable prognostically and since rest possibly is beneficial during acute illness, muscle testing and other examinations are limited to measures necessary for diagnosis and are then confined to evaluating respiratory, circulatory and electrolyte status.

Treatment of the acutely ill patient is designed to achieve comfort unless specific indications of a problem in fluid balance, urination, respiration or circulation exist Pain is alleviated as much as possible by aspirin and other non-narcotic analgesics At times, intravenous injections of calcium gluconate and aminophylline have been strikingly successful in alleviating pain in acute neuromuscular illness although their mechanism of action is not known Narcotics such as codeine, demerol and morphine are *absolutely withheld* until at least a week after the acute illness has passed They depress neural functions and are therefore hazardous in acute nervous system disease Fatalities have closely followed the administration of narcotics during acute and early convalescent poliomyelitis Barbiturates are similarly prohibited since they may further depress an already diseased respiratory center.

Hot packs are used to greater or lesser degree, depending on the number

tilation and observations may be taken as often as every hour in rapidly advancing paralysis. In an adult a fall in vital capacity to less than 1,500 cc. is regarded as evidence of impending respiratory failure, and a fall in vital capacity to 1,000 cc. or less indicates the probable need for artificial respiration. In children, correspondingly lower values are used as indicating a need for artificial respiration and are calculated as approximately one-fourth of the expected normal vital capacity.

Chemical Observations

Most patients with acute neuromuscular diseases fail to develop any significant change in blood or urine chemical values or the body electrolyte, although marked alterations do occur during the illness in a few patients The number and type of desired blood chemical determinations depend on whether the patient is eating, whether he is vomiting, and whether he has respiratory insufficiency with acidosis. Some of the following determinations are only rarely necessary, but all may be needed to guide treatment in an occasional patient:

1) The venous plasma carbon dioxide combining power is ascertained on every patient soon after his admission to the hospital so as to give a base line to which subsequent changes can be compared. If spirometric determinations reveal no significant reduction in ventilation the analysis is not repeated, but if vital capacity falls below 1,500 cc., repeated CO_2 combining power values are determined at intervals of twelve hours or less Progressively increasing values of CO_2 combining powers in the presence of an acid urine mean CO_2 retention and respiratory acidosis which demand correction by increasing ventilation.

2) Blood urea nitrogen (or non-protein nitrogen) is usually obtained. Few persons suffering from the acute neuromuscular diseases, have any defect in urea excretion However, since many require parenteral fluid therapy, including potassium salts, assurance that no toxicity will occur is increased by a knowledge of the state of renal function.

3) Serum protein with albumin-globulin fraction is determined on admission and repeated at regular intervals if the food intake is impaired. Serum proteins may become depleted in any acute illness, and this depletion is accentuated when food intake is limited Bower and his group (5) claimed that in poliomyelitis dramatic changes in serum proteins may occur and have an unfavorable influence on the illness if not prevented or immediately corrected. That this phenomenon is regularly or even frequently seen in human poliomyelitis has not been confirmed. Though plasma probably is not indicated for routine treatment, depleted serum proteins are an indication for parenteral protein replacement when dietary intake is reduced.

The patient is left undisturbed if no reduction of vital capacity below 1,000 cc occurs, and if no sign of respiratory obstruction, hypoxia or hypercapnia appears. On the other hand, if the clinical condition continues to deteriorate, if the vital capacity falls below 1,000 cc., if the CO_2 combining power shows a steady rise, or if signs of deep pulmonary obstruction appear, immediate measures are taken to correct the situation and restore respiratory sufficiency.

Procaine penicillin, 400,000 units daily, is given to all patients with swallowing difficulty or significant respiratory distress to reduce the likelihood of pulmonary infection.

The decision to use a respirator in an adult usually is made when his vital capacity falls to less than 1,000 cc. For acutely ill patients, the tank respirator is always used to provide artificial respiration

Before putting a patient in a respirator a rapid but fairly complete evaluation of the factors contributing to his respiratory distress is made so that if a tracheotomy is indicated it can be performed before artificial respiration is started If he has paralysis of swallowing, total laryngeal paralysis, or is in the first 24 to 36 hours of progression of his paralytic illness, it is assumed that serious obstruction to his airway has already occurred or is extremely likely to occur. Obstruction is assumed in a stuporous or comatose patient or in one with an intrapulmonary infection. With manifestations of any of these conditions, a tracheotomy is performed immediately. A preliminary bronchoscopy is often performed to insure the airway. The tracheobronchial tree is then cleaned of secretions as completely as possible with deep bronchial suctioning. If the clinical condition of the patient and his ventilation do not improve immediately artificial respiration by a tank respirator is instituted. This methodical preliminary evaluation before using artificial respiration is possible only when the failure of ventilation is recognized early If the decision to use artificial respiration is delayed until the patient is in desperate need of air a careful search for obstruction will often be impossible In such circumstances the mortality rate increases sharply

Patients with respiratory distress who have no signs of obstruction and who fail to demonstrate any of the usual manifestations which warn of impending occlusion of the airway receive direct laryngoscopy before being placed in a respirator This allows relatively complete visualization of the larynx and upper trachea, yet does not excessively stimulate protective secretions. If the airway is clear they are then placed in a tank type respirator Actually only a few patients represent a sufficiently simple problem in artificial respiration to justify being put in a respirator without a tracheotomy.

Bronchoscopy not followed by tracheotomy is rarely performed in the acutely ill because bronchoscopy itself educes intense tracheobronchial

of nurses available, but for acutely ill patients they produce little relief of pain after the period of actual use That hot packs serve any function other than to make patients comfortable is doubtful

The Management of Respiratory Failure

If vital capacity falls below 1,500 cc. in adults (and to correspondingly lower values in children) oxygen therapy is begun with a nasal catheter and an oxygen supply of 4 liters per minute Water cylinders through which the oxygen passes for humidification are heated so as to warm the mixture and increase its vapor content. A cold, dry stream of oxygen causes headache and nasopharyngeal irritation with an increase in secretions. Nasal catheters are lubricated with a water-soluble jelly, and changed every four hours to avoid the formation of excessive crusting and adherence to the nasal mucous membrane Oxygen, under these circumstances, is used to reduce fatigue in patients who are still in respiratory compensation. It is not expected to supplant artificial respiration if such is indicated and may actually be dangerous in states of underventilation if the threat of hypercapnia is neglected When oxygen therapy is started, therefore, plasma carbon dioxide combining powers are checked at intervals of 6 to 12 hours because of the increased likelihood of carbon dioxide accumulation when anoxemia is removed as a respiratory stimulant.

If there is difficulty in swallowing or coughing and excessive nasopharyngeal secretion, the patient is placed on his face with a towel beneath his mouth and fluids allowed to drain spontaneously Such patients are also placed in Trendelenberg position 20 minutes out of each hour in an effort to increase drainage from the tracheobronchial tree If the face-down position is used, this degree of tilt should be enough to accomplish gravitational drainage since the trachea has its laryngeal end lower than its bronchial end with the patient prone (21) Intermittent nasal and pharyngeal suction is then applied to keep the airway clear We emphasize *gentleness* with suction pressure, for otherwise the trauma of repeated suctioning stimulates pharyngeal secretions

Endoscopic and respiratory equipment are in immediate readiness near the bedside (though not in view of the patient), and a physician is in constant attendance. The equipment most likely to be needed is always maintained in smooth working order a bronchoscope, laryngoscope, head mirror and laryngoscopic mirror, and an emergency tracheotomy set In addition, a reliable suction machine, several firm rubber whistle-tip catheters (#10, 12, 14), and sterile rinsing solutions are available A rubber or metal oral airway is on hand, and oxygen tanks and humidifiers are present A spare tank respirator is available whenever possible, and an efficient source of bedside lighting is prepared Electrical circuits are checked in advance of using all apparatus, so that fuses will not be overloaded.

5,500 to 6,000 cc. (calculated by taking observed minute volume and subtracting respiratory rate times estimated respiratory dead space).

Occasionally patients are unable to open their glottis synchronously with the negative pressure phase of the respirator. This phenomenon is particularly likely when the respiratory center is involved by poliomyelitis or with delirium or excessive drowsiness. In the presence of a closed glottis the respirator is completely ineffective and, if synchrony cannot be induced within a few minutes by the coaching efforts of the physician, the glottis must be bypassed by immediate tracheotomy. Less vigorous measures than the creation of an artificial airway under these circumstances usually are unsatisfactory for hypoxia develops promptly.

Ventilation is measured in tracheotomized patients just as in others. A small tube is connected between the tracheotomy and spirometer, and readings taken while the nose and mouth are held closed. With a tracheotomy, the estimated respiratory dead space is reduced by approximately one-third in calculating alveolar ventilation.

Postural drainage with the respirator in Trendelenberg position is instituted almost immediately since normal cough mechanisms have long since been lost. Because the head-down position has undesirable effects on ventilation, circulation and intracranial pressure, Trendelenberg position is usually used a maximum of 15 to 30 minutes out of each hour. When first using drainage, the foot of the respirator is raised slowly so as not to evoke nausea, syncope and other effects of sudden changes in intracranial vascular dynamics. Ten to fifteen minutes are allowed in raising the patient to drainage position for the first time.

Nausea and anorexia are frequent in these acutely ill patients. Learning to swallow in phase with the respirator is difficult Because of the danger of inhaling pharyngeal contents, intravenous feeding is used until patients become accustomed to the respirator's action or until they are able to come out of the machine for brief periods Swallowing small amounts of water is tried several times before oral feeding is resumed Despite the increased caloric demands of acute infection, tube feeding is not generally instituted for at least several days Patients being tube fed in respirators often develop gastric distention, nausea and vomiting The danger of inspiration of vomitus is thought to outweigh the desirability of a high caloric intake during this stage. Three grams of potassium chloride and five grams of sodium chloride are given daily in parenteral fluids to prevent depletion. Less viscosity and difficulty in the removal of secretions is observed if 2,000 to 3,000 cc of fluids are given daily. If large amounts of pharyngeal or tracheobronchial secretions are being removed, commensurately greater fluid intakes are supplied

Position is changed frequently to avoid decubitus ulcerations, improve

mucus secretions and provision must be made for their subsequent removal.

Motor power, tank pressure, rubber arm and neck collars, and foot supports are carefully checked on each respirator before being used for artificial respiration.

Being placed in a respirator is a terrifying experience for most patients and should be attended by as little fuss and confusion as possible Whenever time permits, which it usually does if the patient has been evaluated carefully throughout his illness, infusions are started before transfer from bed to respirator. The physician who has had the closest contact with the patient and has inspired his confidence, directs the preparation for artificial respiration with as few assistants as are necessary to perform the task. The need for each step is carefully explained to the patient, who is made as comfortable as possible in the respirator with foot boards supporting his feet, a small pillow placed underneath his knees, and his legs supported in slight internal rotation. A pad is inserted under the small of the back to provide support and prevent pain. Gauze rings are placed beneath the heels to prevent pressure necrosis, and foam rubber pads are inserted between the top of the shoulders and the head of the respirator to soften localized pressure if Trendelenburg drainage is to be used Soft flannel wrappings are placed around the neck to prevent skin abrasions from the respirator collar Gastric dilatation commonly develops with paralytic respiratory insufficiency and may interfere with ventilation If such dilatation exists, the stomach usually is aspirated by Levine tube, in order to avoid interference with breathing and to reduce the likelihood of vomiting.

After these preparations, the respirator is closed If a tracheotomy tube is in place the respirator collar is depressed below the tube by heavy wire held beneath the collar plate to prevent agitation of the fresh wound

With emergency endoscopy equipment available to relieve any sudden block of the airway, respirator pressures are turned on and gradually increased. The physician sits at the patient's head, coaching him in the initially difficult task of synchronizing his feeble breathing efforts with the cycling of the tank Respirator pressure is gradually increased to a level of about $-15 + 5$ cm. H_2O intratank pressure, and the patient encouraged to relax and let the respirator "breathe" for him After a moderate period of time to allow this relaxation and synchronization, ventilation is measured with the spirometer. Respirator pressures are then adjusted so as to achieve tidal volumes of 450 to 550 cc and the respirator rate is adjusted to 16 to 18 cycles per minute. If lower tidal volumes are the best that can be obtained, despite the use of maximal respirator pressure (25 to 30 cm. water negative and 5 to 10 cm water positive pressure), the rate is set correspondingly higher so as to achieve a minute alveolar ventilation of about

THE CARE OF PATIENTS WITH TRACHEOTOMIES

Tracheotomy tubes require special consideration. Although many nurses have had the experience of caring for tracheotomized patients, few are skilled in nursing the individual who has not sufficient expiratory force in his chest to bring pulmonary secretions up the the orifice of the tracheal tube for removal. Extremely deep tracheobronchial suction must be used on patients with paralytic respiratory insufficiency to keep even the larger tracheobronchial passageways patent. If such deep suction is not done gently, recurring tracheobronchial edema counteracts any advantages gained by removal of free secretions. Because of this, and because too enthusiastic suctioning is followed by blood tinged aspirate, no apprentice on the Cornell Service gives tracheotomy care until thoroughly instructed by an experienced nurse or physician. Before suction is turned on, the tracheal catheter (a # 10–12 whistle tip) is advanced into the tracheotomy and to the full depth of the right main stem bronchus (the right is the path of least resistance). The catheter is then withdrawn about a half-centimeter to prevent it "grabbing" against mucous membrane of the lower bronchus and negative pressure turned on. When suction of secretions becomes audible (5 to 10 sec) the catheter is withdrawn slowly and twisted all the way out. If the catheter sticks against the wall of the tracheobronchial tree, suction is immediately turned off—in this way pieces of membrane will not be pulled away. Total time for withdrawal of the sucking catheter is about 30 seconds since prolonged deep tracheobronchial suction causes hypoxia, as first shown by Elam and his colleagues (1948) Catheters are *never* jabbed back and forth down the trachea as this traumatizes the membranes. Even if all the secretions are not removed, 2 to 3 minutes are allowed for the patient to re-oxygenate and rest before the procedure is repeated. When the right main stem bronchus has been cleaned, the tracheotomy tube is gently tilted to the left, usually firmly enough for the operator to feel transmitted aortic pulsation, and the catheter passed into the left main stem bronchus. The same procedure is then repeated. Suctioning is repeated as often as indicated, usually every ½ to 1 hour during the acute stages of illness Catheters are kept clean in 5 per cent sodium bicarbonate solution, and clearly marked so that nasal and tracheal catheters are separated If pulmonary secretions are quite viscous, ½ cc of 5 per cent sodium bicarbonate solution is instilled into the tracheotomy tube immediately before suctioning This tends to reduce the tenacity of the secretions.

Outer tracheotomy tubes usually are not replaced for 48 hours following their initial introduction, so as to avoid hemorrhage. Usually they are changed daily thereafter during the acute illness The inner cannula is removed, cleaned and re-inserted at least twice a day for the first few days

systemic circulation and assist pulmonary drainage. Respirator patients are turned every hour when awake and every two hours when sleeping To prevent the inside of the respirator from becoming excessively hot, the inside light is left off All respirators have inside thermometers, and on particularly warm days ice is suspended on rubber sheeting across the inside of the tank and air circulated with a small inside fan.

Frequent checks are made of ventilation. Spirometer recordings are made at least once daily, more frequently in the seriously ill. Oxygenation is indirectly checked by evaluating skin color and pulse rate and occasionally oxygen saturation is directly analyzed by oximeter or blood gas analysis. Carbon dioxide combining powers are repeated at 12 to 24 hour intervals. The chest is auscultated at regular intervals to detect bronchitis, pneumonitis or atelectasis An indwelling stethescope connected through a rubber cork in the respirator head piece eliminates the difficulty of trying to listen through the arm portals and permits more accurate observations. The point of maximum impulse of the heart is usually marked with ink at the time of placing a patient in the respirator since a change in position of the PMI may be the earliest sign of atelectasis. If hypoxia or hypercapnia cannot be reversed by suctioning and the use of maximal respirator pressures, oxygen is administered under intermittent positive pressure synchronized with the inspiratory cycle of the respirator This is usually applied alternately with rest periods since it is somewhat uncomfortable and often induces claustrophobia. The schedule adopted depends on the patient's ventilatory demands, but one frequently used is ten minute breathing periods alternating with five minute rest periods.

If tracheobronchial obstruction, atelectasis or intensive pneumonitis develop in an acutely ill respirator patient who does not already have a tracheotomy, bronchoscopy followed by tracheotomy is usually performed and occasionally must be carried out in a few seconds if the patient's life is to be saved. The procedure in such emergencies is to pass the bronchoscope immediately and remove any existing obstruction in the main stem bronchi, leaving the patient in the respirator during this maneuver With either the bronchoscope or an endotracheal tube *left in place*, positive pressure breathing is started with a closed system anesthesia unit, using manual compression of the reservoir bag. The patient is removed from the respirator and a tracheotomy performed over the endotrachial tube He is then returned to the respirator, the respirator collar is adjusted so as not to chafe the tracheotomy wound, and the endotracheal tube and positive pressure breathing unit are removed Artificial respiration is then resumed by tank respirator.

On the Cornell-New York Hospital Service most patients with ventilatory insufficiency are not removed from tank respirators until after acute illness has passed and their temperature has returned to normal or their signs of neurological dysfunction have begun to recede. Occasional trials of independent breathing with the respirator turned off may be tried cautiously before this, unless the patient becomes so panicky as to interfere with his readjustment to the respirator. Though reliable data as to tidal volume and vital capacity are desirable, usually it is safe to rely on the statement of the patient as to his ability to breathe without the respirator. Patients do not rapidly develop significant hypoxia or marked hypercapnia without symptoms. As a corollary, many patients who when superficially evaluated are considered "psychologically dependent" on respirators actually have physiological insufficiencies which need artificial respiration. To press such individuals to remain out of respirators for excessive periods may have disastrous results. However, patients in whom adequate ventilatory function can be demonstrated occasionally express considerable apprehension about being removed from respirators. They usually overcome their fears quickly when their dependent needs are recognized and they are carefully coached in breathing by a physician in whom they have confidence One must know the amount of breathing of which such patients are capable, for if their respiratory demands exceed their capacity they become terror-stricken and their recovery is seriously delayed.

When acute illness has passed, brief trials of independent breathing without artificial respiration are started with the patient still lying in the respirator. First this is done with merely an arm port open, later with the respirator carriage partly extended Measurement of ventilation at this stage is not attempted unless breathing can be maintained independently for five minutes or so, for the entire concentration is usually given to the respiratory efforts and accurate records are impossible Some will be able to breathe for a considerable time even at this first trial but if it is demonstrated that independent ventilation is possible for only a few minutes or seconds practice periods of breathing 3 to 4 times a day are instituted Records of the duration of each of these spontaneous breathing periods are made and charted by the bedside to show patients the progress of their efforts. An attempt is made to prolong the time out of the respirator each day, even though the increase may be only a matter of seconds.

Convalescent patients who are able to breathe for only a few minutes or seconds are placed in cuirass respirators as soon as possible Since a cuirass produces a distinctly different feeling of breathing, and usually results in lower tidal volumes than the tank, preliminary short trials with a cuirass respirator are made while the patient is still on the cart of his tank respirator. As his adjustment improves, the tidal volume is determined and re-

and the obturator is inserted into the outer cannula at these times to insure its patency. It is especially important to avoid plugging the lower end of tracheotomy tubes by dried and crusted secretions.

At times tracheotomized respirator patients develop signs of gross atelectasis or pneumonitis despite repeated attempts to clean out the tracheobronchial tree by suction. Such pulmonary complications more frequently develop in the left lung because of the anatomical difficulty in reaching the left main bronchus. Bronchoscopy usually reveals the obstruction responsible for such lesions and assists in its resolution. If atelectasis develops on the right, bronchoscopy is less imperative for there are few places a bronchoscope can reach that cannot be drained by blind suctioning through the tracheotomy. Most patients with a tracheotomy who develop a right sided atelectasis have their obstruction distal to the main stem bronchus and are likely to be more helped by placing them on their left side and pounding the chest so as to loosen the secretions. Bronchoscopic examination is thus usually withheld for right lung consolidation except to identify which segment of the lung is most involved. Used in this way, the proper position for drainage may be established

A tracheotomy considerably eases the technic of bronchoscopy since the instrument can be passed directly through the tracheal stoma. Using this technic, bronchoscopy has been done three to four times a day without excessive trauma in relieving a refractory left-sided atelectasis.

The acute stage of paralytic respiratory insufficiency is managed best by artificial respiration begun early and tracheotomy performed before significant obstruction develops With barbiturate or morphine poisoning, a respiratory emergency rarely lasts more than a few days, while with poliomyelitis or polyneuritis the acute fulminating period of respiratory insufficiency is usually over within two weeks. In myasthenia gravis the duration of ventilatory failure is unpredictable and usually depends on whether response to prostigmine returns Of all these illnesses only polyneuritis and poliomyelitis, particularly the latter, require extensive convalescent respiratory rehabilitation

Management of the Respirator Patient When the Acute Stage Has Passed—the Recovery Phase

Many patients require continuous artificial respiration for only a few hours and suffer little or no permanent impairment of ventilation Others may be left with so little residual breathing musculature as to be at least partly dependent on mechanically induced respiration the rest of their lives. Since most fall between these extremes a careful program of convalescent care speeds recovery and in a few instances may be the deciding factor in re-establishing independent breathing.

most instances, even when artificial respiration is needed for only a few hours out of each 24, patients use tank respirators at night. This selection is made because these respirators produce greater tidal volumes for comparable increments of pressure, and therefore increase chest expansion and

TABLE 1

Vital Capacities Attained by Twenty Patients Convalescing from Extensive Neuromuscular Disease (Poliomyelitis) with Respiratory Insufficiency

Predicted vital capacity based on body surface area in sq meters × 2 5 for males and × 2 0 for females Convalescent values recorded at earliest time patients were able to breathe independently for the stated periods

PATIENT	AGE	SEX	VITAL CAPACITY		
			Expected normal	Able to breathe without respirator one hour	Able to breathe without respirator completely
	yrs		cc	cc	cc
M. T	18	F	3,300	550	850
J R	26	F	3,000	300	735
E S	25	F	3,000	530	620
I P.	18	M	4,650	490	820
J F	21	M	4,850	365	550
G H	23	M	4,650	215	575
O H	18	M	4,700	395	460
E C	16	M	4,200	490	760
M D	33	M	4,600	390	610
J B	24	F	3,000	330	365
G R	16	M	4,700		600
F C	6	M	1,600	230	550
J T	14	F	3,400	581	700
P W	18	M	4,700		850
H H	29	M	5,100	610	915
E L	31	F	2,900	518	1100
J L	21	F	2,900	550	650
R S	28	M	4,750	320	
D B	22	M	4,900	800	1100
J W	23	M	4,300	350	500

keep the thorax flexible Every effort is made to maintain the chest as easily expansible as possible so that less intrinsic effort will be required if muscular control of breathing returns By using rocking beds and cuirass respirators as early as possible in convalescence, weaning from tank respirators is now much easier than in former years, and the number of chronic respirator patients appears to have been considerably reduced

It is possible for patients to remain out of the respirator for short periods even when the vital capacity is remarkably low As noted in table 1, most

spiratory rate adjusted to produce adequate minute volume. He is then transferred to a bed with the cuirass in place. Baths, enemas and physiotherapeutic exercises are carried out during these periods, for the extremities and back are free and easily attended to. Patients are gradually brought to a sitting position and if the amount of muscular power in their extremities is proportionately greater than their ability to breathe, they are taught to brush their teeth, wash and eat with a cuirass in place. Those with more or less permanent severe limitations of breathing are eventually gotten up in wheel chairs with the cuirass. Progressive breathing exercises are continued each day in an increasing effort to re-achieve independent breathing.

As soon as patients are able to ventilate without assistance for five minutes or more they are usually transferred to a rapid rocking bed. The rate of rocking is at first made sufficiently rapid to eliminate subjective sensations of air hunger, but as soon as possible ventilatory measurements are made and the rate adjusted to produce about 5,000 cc. of minute alveolar ventilation. With standard commercial rocking beds which rock through total arcs of 40 to 44 degrees, the maximal rocking arc is always used, to give the greatest tidal volume Periods of rocking are at first limited to a few minutes to avoid fatigue, then gradually increased from one-half to one hour or more. Most patients comfortably enjoy rocking for as much as 4 hours within two weeks after acute illness has passed In the event that independent breathing is not sufficient to support respiratory demands long enough to effect transfer from tank respirator to the rocking bed, a cuirass respirator is applied while moving the patient and making him comfortable. The rocking bed is then started up and the cuirass removed for a few minutes In this way, gradual adjustment to the new experience of rocking is made possible

As soon as the patient is sufficiently accustomed to rocking to be ventilated comfortably for half an hour or more, nursing and physiotherapeutic care is administered during rocking No encumbering apparatus is on the body so these procedures can be more efficiently applied at this time and, in addition, the patient can be turned on his face so that proper attention can be given to his back At the same time, diaphragmatic fixation is prevented by the intermittent pressure of the abdominal viscera.

The daily routine for these convalescent patients is a busy one. For those who are improving rapidly in their ability to breathe without artificial respiration, periods of independent breathing are usually used first for eating, and other activities are added later to their respirator-free times For individuals who need nearly constant artificial respiration far into convalescence a daytime routine of using a cuirass respirator during feeding and bowel movements is usually employed, and much of the rest of the day is spent enjoying the comparative freedom of the rocking bed In

spontaneously and heal by granulation. This spontaneous closure is achieved within as little as 24 hours in some cases.

Every effort is made to prevent respiratory infections during recovery from paralytic respiratory insufficiency by carefully keeping these patients from contact with persons who have colds. If a cold develops in spite of these precautions it receives immediate attention, for an apparently mild rhinitis may quickly develop into a severe bronchitis or pneumonitis when coughing mechanisms are lost or inhibited Also, as Lukas has demonstrated, respiratory infections clinically limited to the nose and throat significantly impair vital capacity and maximal breathing capacity even in normal persons. In pre-antibiotic years Landon (30) and Brahdy (7) noted that such infections caused a high mortality among those recovering from paralytic respiratory insufficiency. Even though anti-bacterial therapy and the widespread use of tracheotomy have now made such deaths rare, the possibility of chronic bronchitis, bronchiectasis and emphysema cannot be ignored. At the first manifestation of rhinitis, pharyngitis or bronchitis, nose and throat exudates are cultured and penicillin or aureomycin given prophylactically. The appropriate anti-bacterial agent is then selected when culture results are known. In addition to anti-biotics, a broncho-dilator, such as a vapo-nephrin spray, is used at four hour intervals since bronchospasm commonly accompanies upper respiratory infections and further impairs the already limited ability to ventilate. Postural drainage in Trendelenberg position is instituted with the patient face down on the bed for 10 to 15 minutes out of each hour unless an effective cough is present Gentle nasopharyngeal suction removes secretions and prevents drainage into the lungs. The lungs are examined repeatedly, for obstruction of the respiratory passageways or pneumonitis. A recently closed tracheotomy may need to be re-opened when bronchitis is severe. Bronchoscopy is resorted to in convalescence more frequently than during the acute paralytic illness in the treatment of obstruction, for respiratory function is expected to improve rapidly after the transient infection, and fresh tracheotomy may not be necessary. Ventilation is measured repeatedly, as during the acute paralytic illness, for it is apt to diminish during infection and re-application of the respirator may be necessary.

Convalescence following a respiratory infection in which deterioration of respiratory function has taken place sometimes requires reinstitution of the same regimen used in the acute paralytic illness These patients are often so debilitated that slow progress is the best that can be hoped for.

Close attention is given to the nutritional status of those recovering from acute neuromuscular illnesses It is not unusual for an adult who is extensively paralyzed to lose thirty or forty pounds in weight. Although tube feeding is contraindicated during the acute illness because of gastric atony,

adults convalescing from paralytic respiratory insufficiency are able to be partially independent of the respirator when their vital capacity comes up to 300 to 500 cc. and completely independent when their vital capacity reaches 600 to 800 cc. Ten to fifteen minute breathing periods may be accomplished with vital capacities as low as 175 cc.

In addition to being placed in a cuirass respirator and/or transferred to the rocking bed, each convalescent patient also performs exercises of blowing against progressively increasing resistance, either by inflating balloons or blowing increasing amounts of fluid from one flask to another through a rubber tube. This exercise attempts to gradually improve expiratory force. If a considerable degree of fixation of the chest has occurred, due to replacement of muscle by fibrous tissue, the chest is inflated passively by positive pressure breathing.

All patients with paralysis of respiratory muscles are instructed to breathe as rapidly and deeply as possible for two minutes of every hour. It is hoped in this way to increase their maximal breathing capacity and breathing reserve Since ventilation depends on rate of breathing nearly as much as depth of breathing, some patients who have been in respirators for long periods of time actually have to be trained in this way to breathe rapidly again.

The vital capacity is measured at weekly intervals up to the time of discharge from the hospital or until stabilized at a constant level for several weeks Vital capacity measurements are continued less frequently for many months after apparent stabilization since increases of 20 to 30 per cent are seen occasionally even six or more months after the acute illness.

Tracheotomy tubes are usually left in place until the vital capacity rises to between 1,200 and 1,500 cc , for with volumes below this level cough remains ineffective. Without ability to cough, sporadic respiratory infections may obstruct the tracheobronchial tree and necessitate reopening a closed trachea Ocasionally, if only a small amount of the breathing ability has returned after several months' convalescence, tracheotomy tubes are removed despite lower vital capacities This is done only when further ventilatory improvement seems unlikely.

As a preliminary to being removed, or when tracheobronchial suction is no longer necessary, tracheotomy tubes are usually corked to allow speaking and practice in coughing and subjective clearing of the throat. For patients who have such a limited ability to breathe that total removal of the tracheotomy is unwise, the corked tube causes little discomfort and indeed its presence is eventually practically ignored. For those with better recovery of breathing ability, corking the tube allows a fairly reliable prediction of whether the tracheotomy can safely be closed permanently. When finally removing the tracheotomy, the stoma is allowed to close

be associated with a lower vital capacity as a result of intercostal muscle paralysis.

It is best to measure maximal breathing capacity, vital capacity and the movement of the chest under fluoroscopic examination before discharge from the hospital. Lukas and Plum (34) found that the combination of vital capacity and chest fluoroscopy gives the required information, if the fluoroscopic examination includes examination of the thoracic wall and diaphragms during breathing performed as deeply and rapidly as possible.

Patients are discharged from the hospital after acute paralytic respiratory insufficiency when their breathing has returned sufficiently so that no artificial respiration is needed, and they have the necessary breathing reserve to lead their regular life without undue exhaustion. This demands the removal of tracheotomy tubes. Actually, since acute poliomyelitis and polyneuritis are the most common illnesses responsible, decision for discharge from hospital usually depends on the return of neuromuscular function in the extremities, and the need for treating residual paralysis in these regions. It is rare for a patient to walk well enough to qualify for out-patient care and still have enough breathing paralysis to need hospitalization.

Individuals who have been discharged from the hospital with any significant residual limitation of breathing are re-examined at intervals of one to three months in the out-patient department Their activity gradually increases as their ventilatory function improves The need for continued effort to develop rapid and deep breathing is stressed Such patients report to the hospital immediately with any respiratory infection, and if any difficulty in dealing with the infection is apparent they are admitted for closer supervision. By establishing this close relationship between out-patient and in-patient supervision, adult patients have been cared for excellently. Many have been discharged from the hospital with vital capacities as low as 700 to 1,000 cc.

THE AIM OF TREATMENT IN NEUROMUSCULAR RESPIRATORY FAILURE

Although this monograph has been limited to the actual physical care of the patient's breathing apparatus, it should be stressed that motivating all the procedures, tests and treatments is the fundamental goal of re-establishing a useful human existence Far more than physical measures are necessary to achieve this goal, and social, vocational, sexual and emotional re-adjustment must be sought commensurately with breathing exercises Within four to six weeks after poliomyelitis and polyneuritis, and even earlier in other neuromuscular illnesses, a reasonably close estimate can be made of the degree of eventual recovery Using this estimate, the patient can be prepared for the modifications on his life that his illness will im-

or nausea and vomiting, it is started early in convalescence for those with paralysis of swallowing or persistent anorexia. Since anorexia may persist for many weeks after these illnesses, particularly after poliomyelitis, special efforts are made to provide appetizing food. For some, dividing meals into smaller and more frequent feedings enables the caloric intake to be increased. A dietary intake of 2500 to 3,000 calories daily is the goal If prolonged recumbency is likely, the calcium intake is kept at moderate levels to reduce the likelihood of renal calculi Fluid intake is maintained between 3,000 to 3,500 cc. for the same reason The management of the patient in whom ambulation does not take place within the first month has recently been reviewed by Whedon (65).

As ventilation improves, patients are given progressively more difficult exercises to develop their breathing. At first by passive later by active movement, they are encouraged to exert themselves and put increasing demands on their respiratory abilities. When the vital capacity reaches 500 to 600 cc. an attempt is made to prolong the periods out of the respirator into the evening, and patients are encouraged to take short naps without artificial respiration Finally the period out of the respirator is extended to an entire day and night. If the patient cannot sleep he is not sedated, for insomnia may be a protective device against insidiously developing hypoxia or hypercapnia. A central nervous system depressant is not used to induce sleep until patients are able to breathe permanently using their own muscles. When this stage is reached in adults the vital capacity will almost invariably be greater than 700 to 800 cc.

EVALUATION OF RESPIRATORY FUNCTION PRIOR TO DISCHARGE FROM TREATMENT

Vital capacity determination provides a simple and valuable comparative index of the state of ventilation in the acute and convalescent stages of paralytic respiratory insufficiency It must be supplemented by other observations for a complete analysis of the amount and type of breathing function remaining after maximal improvement has occurred This is as true with only moderate impairment of breathing where artificial respiration was never needed as with severe respiratory involvement The rapidity with which air can be moved in and out of the chest and the function of the diaphragm must be known in order to have a clear idea of the ability to tolerate exercise and withstand infection Unilateral paralysis of the diaphragm, for example, may cause relatively little reduction in vital capacity, but because it leads to impaired drainage of basal pulmonary segments on the same side it may lead to chronic bronchitis or bronchiectasis. Respiratory infection in such instances requires more care than when bilateral diaphragmatic function is intact, even though the latter might

are surprisingly good and effectively dispel any notions that the requirement of a respirator for an acutely ill person implies a future life of invalidism. Table 2 lists the present activities of our adult patients cared for in respirators in the past three years. It is evident that though some have been unable to return to active life, they represent, as a whole, a group of far from helpless individuals

ACKNOWLEDGEMENT

In preparing a monograph of this type, it is impossible to give due credit to all those who have helped shape one's thoughts and ideas on the subject. However, deep appreciation is expressed to those colleagues and members of the staff of Cornell University Medical College with whom the author has had the privilege of being associated during the three years that the material presented has been under investigation A particular indebtedness is due Drs. D S Lukas, G. D. Whedon, F. H. McDowell and F. T. Kirkham, in association with whom much of the work was performed. Dr. Harold G. Wolff has been a constant source of inspiration and constructive criticism. Finally, the entire project would have been impossible without the dedicated efforts of many of the nurses of the Department of Medicine of the New York Hospital.

REFERENCES

1 Asmussen, E. and Neilsen, M . Efficacy of artificial respiration J Appl Physiol , 3: 95, 1950
2. Baldwin, E de F , Cournand, A. and Richards, D W Pulmonary insufficiency I. Classification, methods, and standards Medicine, 27: 243, 1948
3 Barach, A L , Fenn, W O , Ferris, E B and Schmidt, C F The physiology of pressure breathing A brief review of its present status J Aviation Med , 18: 73, 1947
4 Behnke, A. R Certain physiological principles underlying resuscitation and oxygen therapy. Anesthesiology, 2: 245, 1941
5 Bower, A G , Chaney, A L , Eaton, R M , Chudnoff, J and Affeldt, J Patterns of plasma protein changes in acute poliomyelitis patients treatment with blood plasma Am J Med Sci , 220: 46, 1950
6 Bower, A G , Bennett, V R , Dillon, J B and Axelrod, B . Investigations on the care and treatment of poliomyelitis patients Ann West Med. & Surg , 4: 686, 1950
7 Brahdy, M B Late results of 63 cases of poliomyelitis treated in the respirator. N Y State J Med , 36: 1147, 1936.
8 Brazier, M A B · Physiological effects of carbon dioxide on activity of central nervous system in man with special reference to problem of high altitude flying review Medicine, 22: 205, 1943.
9 Brown, E W Physiological effects of high concentrations of carbon dioxide U S Nav M. Bull , 28: 721, 1930
10 Bruns, O : Über Weiderbelebung durch Kunstliche. Atmung, Klin Wchnschr , 6: 1548, 1927.

pose. Occupational therapy, instead of being a diversional tool, can play an active part in this human re-creation, and social agencies can lend their valuable resources so that when the patient leaves the hospital he will find he still has a world in which to live and work. The results of such a program

TABLE 2

The Socio-economic Activity of Twenty-two Patients Two Years or More Following Acute Neuro-muscular Disease

Each of these patients required artificial respiration to survive the acute illness

PATIENT	AGE	SEX	DURATION OF ARTIFICIAL RESPIRATION	TOTAL DURATION OF TREATMENT	PRE-ILLNESS OCCUPATION	PRESENT OCCUPATION	ESTIMATED DISABILITY
							%
F. C	7	M	2.5 mo.	2 5 yr.	Grammar school	Special grammar school	60
O S	10	M	2.5 yr.	2 5 yr	Grammar school	Home patient	95
J O'R.	10	M	10 da	4 mo.	Grammar school	Grammar school	0
G R	15	M	16 da	9 mo	High school	High school	30
J T	15	F	15 da	9 mo	High school	High school	10
E. C	18	M	2 mo	10 mo	Handyman	Unemployed	20
I. P	18	M	21 da	1 5 yr	High school	College	25
P W.	18	M	3 da	6 mo	High school	Clerk	10
O H	18	M	1 5 mo	1 5 yr.	High school	Home patient	90
J F.	20	M	2 mo	1 5 yr	Bank runner	Hospital patient*	50
D. B	21	M	20 da.	4 mo	Engineer student	Mechanical engineer	10
J. L.	21	F	36 da	8 mo	Housewife	Housewife	25
G H	23	M	2 yr	2 yr	Mechanic	Hospital patient	95
J B.	25	F	28 da	1 yr	Housewife	Housewife (limited)	60
E. S.	26	F	4 mo	1 5 yr	Housewife	Housewife	25
H H	29	M	3 mo	1 5 yr	Mechanical engineer	Hospital patient*	65
E. L.	32	F	5 da.	4 mo	Housewife	Housewife	10
M. L	34	M	3 yr	3 yr	Lawyer	Hospital patient	100
M. D	34	M	2 5 mo	1 5 yr	Salesman	Accountant	50
A V.†	24	F	19 da	6 mo	Housewife	Housewife	0
D C.†	27	M	11 da.	4 mo	Musician	Musician	0

* Expected to return to active self-sustaining life

† These two patients suffered the Guillian Barré Syndrome, the remainder had acute poliomyelitis.

32 LEAKE, C. D. AND WATERS, R. M.: Anesthetic properties of carbon dioxide Anesth & Analg, 8: 17, 1929
33. LESTER, C W, COURNAND, A AND RILEY, R. L. Pulmonary function after pneumonectomy in children J. Thoracic Surg, 11: 529, 1942
34 LUKAS, D S AND PLUM, F Pulmonary function in patients convalescing from acute poliomyelitis with respiratory paralysis Am J Med., 12: 388, 1952
35 LUNDSGAARD, C AND VAN SLYKE, D D. Cyanosis Medicine, 2: 1, 1923
36 MALONEY, J V. AND WHITTENBERGER, J L: Clinical implications of pressures used in the body respirator Am J Med Sci, 221: 425, 1951
37. McDOWELL, F H. AND PLUM, F: Arterial hypertension associated with acute poliomyelitis New Eng J Med, 245: 241, 1951.
38 MILLER, H. A AND BUCK, L S: Tracheotomy in poliomyelitis Calif Med, 72: 34, 1950.
39 Minneapolis Poliomyelitis Research Commission Bulbar form of poliomyelitis I Diagnosis and correlation of clinical with physiologic and pathologic manifestations J A. M. A, 134: 757, 1947
40 Minneapolis Poliomyelitis Research Commission Bulbar form of poliomyelitis II. Therapeutic measures based on pathologic and physiologic findings J A M A, 135: 425, 1947.
41. MOTLEY, H. L, COURNAND, A, WERKO, L, DRESDALE, D T, HIMMELSTEIN, A, AND RICHARDS, D W Intermittent positive pressure breathing—a means of administering artificial respiration in man J A M A, 137: 370, 1948
42 MOTLEY, H. L, WERKO, L, COURNAND, A AND RICHARDS, D W: Observation on the clinical use of intermittent positive pressure J Aviat Med, 18: 417, 1947.
43 MOYER, C H AND BEECHER, H K Effects of barbiturate anesthesia upon the integration of respiratory control mechanisms J Clin Invest, 21: 429, 1942
44 NEILSEN, H Method of resuscitation Ugesk f Laeger, 94: 1201, 1932
45 NILLSON, E On treatment of barbiturate poisoning Acta Med Scandinav, supplement 253, 1951
46 NIMS, R G, CONNER, E. H, BOTELHO, S Y AND COMROE, J H, Jr A comparison of methods for performing manual artificial respiration on apneic patients. J Appl Physiol, 4. 486, 1951
47 PAGE, I H AND OLMSTED, F The influence of respiratory gas mixtures on arterial pressure and vascular reactivity in "normal" and hypertensive dogs. Circulation, 3: 801, 1951
48 PLUM, F AND LUKAS, D S An evaluation of the cuirass respirator in acute poliomyelitis with respiratory insufficiency. Am J Med Sci, 221: 417, 1951
49 PLUM, F AND WHEDON, G D: The rapid rocking bed: its effect on the ventilation of poliomyelitis patients with respiratory paralysis New Eng J Med, 245: 235, 1951
50 PRIEST, R E, BOIES, L R AND GOLTZ, N F Tracheotomy in bulbar poliomyelitis *Ann Otol, Rhin & Laryng, 56: 250, 1947*
51 Reports on the Use of the Electrophrenic Respirator Cambridge, Mass, The Sanborn Company, 1951
52 ROBINSON, S Experimental studies of physical fitness in relation to age Arbeits physiologie, 10: 3, 1938
53 SARNOFF, S J, MALONEY, J V AND WHITTENBERGER, J L Electrophrenic respiration V Effect on the circulation of electrophrenic respiration and positive pressure breathing during respiratory paralysis of high spinal anesthesia. Ann Surg, 132: 921, 1950.

11. Comroe, J. H., Jr. and Botelho, S.: The unreliability of cyanosis in the recognition of arterial anoxemia. Am J. Med. Sci., 214: 1, 1947.
12 Comroe, J. H., Jr. and Dripps, R. D.: Artificial respiration. J. A. M. A, 130: 381, 1946.
13. Cordier, G G.: Method of artificial respiration. Brit M. J, 2: 381, 1943
14. Council on Physical Medicine and Rehabilitation Back pressure-arm lift method for administering artificial respiration recommended J A. M A., 147: 1454, 1951.
15. Cournand, A, Motley, H L, Werko, L and Richards, D. W. Physiological studies of the effects of intermittent positive pressure breathing on cardiac output in man Am J. Physiol., 152: 162, 1948.
16 Drinker, C. K Pulmonary edema and inflammation: an analysis of processes in the formation and removal of pulmonary transudates and exudates Harvard Monographs in Medicine and Public Health #7. Cambridge, Mass, Harvard University Press, 1945
17 Drinker, P. and Shaw, L A An apparatus for prolonged administration of artificial respiration J Clin Invest, 7: 229, 1929
18 Elam, J O, Hemingway, A, Gullickson, G and Visscher, M B Impairment of pulmonary function in poliomyelitis oximetric studies in patients with the spinal and bulbar types Arch Int Med, 81: 649, 1948
19. Eve, F. G · Actuation of the inert diaphragm by gravity method Lancet, 2. 995, 1932
20 Galloway, T C Tracheotomy in bulbar poliomyelitis J A M A, 123 1096, 1943
21 Galloway, T C and Seifert, M H Bulbar poliomyelitis favorable results in its treatment as problem in respiratory obstruction J A M A, 141: 1, 1949.
22. Goldstein, J D and DuBois, E L Effect on the circulation in man of rebreathing different concentrations of carbon dioxide Am J Physiol, 81: 650, 1927
23 Gordon, A S, Fainer, D C and Ivy, A C Artificial respiration new method and comparative study of different methods in adults J A. M A, 144: 1455, 1950
24 Gordon, A S, Raymon, F, Sandove, M and Ivy, A C Manual artificial respiration. comparison of various methods in apneic normal adults J A M A, 144: 1447, 1950
25 Gordon, A S, Sandove, M S, Raymon, F and Ivy, A C Critical survey of manual artificial respiration J A M A, 147: 1444, 1951
26. Harper, P and Tennant, R · Treatment of respiratory failure in poliomyelitis Yale J Biol & Med, 6: 31, 1933
27 Haymaker, W and Kernohan, J W The Landry-Guillain-Barré syndrome. Medicine, 28: 59, 1947
28 [illegible] P V Hale, C J and Bailey, T L Pulmonary ventilation in [illegible]
[illegible] Physiol,
29 [illegible]
30 Landon, J F An analysis of 88 cases of poliomyelitis treated with the Drinker respirator with a control series of 68 cases, clinical studies in poliomyelitis J Pediat, 5: 1, 1934
31 Lans, H. S, Stein, I F, Becker, R J, Hoyne, A L and Meyer, K A Potassium deficiency in bulbar poliomyelitis J A M A, 146: 1017, 1951

Cortisone and ACTH in Infectious Processes

MAX MICHAEL, JR., M.D.

INTRODUCTION

The profound influence of the secretion of the adrenal cortex on the outcome of an infectious process has long been well-appreciated. In particular, it has been known that patients with adrenal insufficiency tolerate acute infections poorly and are frequently thrown into an Addisonian crisis by a seemingly minor infection (55). When enough cortisone and ACTH became available for clinical investigation, it was only natural to use them in an attempt to modify the outcome of a variety of infections. In some instances, they have been used alone, and in others they have been combined with antibiotics. Changes in existing infections have been noted in patients receiving the hormones for other diseases Germane to this discussion are the reported facts that cortisone and ACTH indeed do alter in an unfavorable fashion the outcome of many infectious processes (13, 30, 46). To understand the mechanisms by which such alterations occur and to use the substances intelligently, it is essential to have some concept of their modes of actions on infections. A mass of clinical and experimental data has been accumulated. It aids in shedding light on some of these observed facts. In view of the present extensive and often indiscriminate clinical use of cortisone and ACTH, these data will be presented and dissected, in an attempt to answer certain practical questions. 1) Do these substances exert any beneficial effect on patients with various infections? 2) Are there infections that are definitely aggravated by these substances? 3) How can the effect on various infections be recognized clinically? At this date, there is a considerable hiatus in our knowledge which must be filled before the posed questions can be answered with finality. This, then, is in the nature of a progress report.

EFFECTS ON CERTAIN DEFENSE MECHANISMS

The entry of an invading micro-organism into the body evokes a chain of events which in the main are detrimental to the invader. In some infections, however, the host's reaction may impede recovery by virtue of accompanying tissue destruction and reparative fibrosis. The out-come of this encounter between invader and defender depends upon: 1) the nature of the body defenses, 2) the virulence of the invader; and 3) the adjunct use of substances inimical to the micro-organisms Soon after a micro-organism

54. SARNOFF, S J., MALONEY, J.V, SARNOFF, L. C., FERRIS, B. G. AND WHITTENBERGER, J. L.: Electrophrenic respiration in acute bulbar poliomyelitis. Its use in management of respiratory irregularities. J. A M. A, 143: 1383, 1950
55 SARNOFF, S J, WHITTENBERGER, J. L. AND AFFELDT, J. E: The hypoventilation syndrome in bulbar poliomyelitis. J. A M. A., 147: 30, 1951
56. SARNOFF, S J, WHITTENBERGER, J L. AND HARDENBERGH, E. Electrophrenic respiration III. Mechanism of the inhibition of spontaneous respiration Am. J Physiol, 155: 203, 1948
57. SARNOFF, S J., SARNOFF, L C AND WHITTENBERGER, J. L: Electrophrenic respiration. VII. The motor point of the phrenic nerve in relation to external stimulation Surg, Gyn & Ob, 93: 190, 1951.
58. SEEVERS, M H. The narcotic properties of carbon dioxide. N. Y. State J. Med, 44: 597, 1944
59 SHAEFER, K. E. (Ed) Symposium on submarine medicine. U. S Fleet Naval Forces, Germany, Technical Section (Medical).
60. SCHAFER, E A Artificial respiration in man Harvey Lectures, 1907–08
61. SHORR, E AND ZWEIFACH, B W. Hepato-renal vasotropic factors in blood during chronic essential hypertension in man Tr Assoc. Am Phys, 61: 350, 1948
62 SJÖBERG, A Mechanism of suffocation in spino-bulbar poliomyelitis and experiences with operative treatment Arch Otol, 52: 323, 1950
63. SWANN, H G Fresh and sea water drowning Medical Division Special Report 5, Conference on Artificial Respiration, p 9, Chemical Corps, Army Chemical Center, Maryland, April, 1951
64. WATROUS, W G, DAVIS, F E AND ANDERSON, B M: Manually assisted and controlled respiration its use during inhalation anesthesia for the maintenance of a near-normal physiological state—a review Anesthesiology, 11: 538–561, 661–685, 1950, 12: 33, 1951
65. WHEDON, G D Management of the effects of recumbency M Clin N. Am, 35: 545, 1951.
66 WHEDON, G D Metabolic studies in poliomyelitis To be published
67. WHITTENBERGER, J L, SARNOFF, S. J AND HARDENBERGH, E: Electrophrenic respiration II Its use in man J Clin Invest, 28: 124, 1949
68 WHITTENBERGER, J L, AFFELDT, J E, GOODALE, W. T AND SARNOFF, S J The mechanics of breathing in relation to manual methods of artificial respiration J Appl Physiol, 4: 476, 1951
69 WILSON, J L Acute anterior poliomyelitis, treatment of bulbar and high spinal types New Eng J Med, 206: 887, 1932
70. WILSON, J L The use of the respirator in poliomyelitis New York, National Foundation for Infantile Paralysis, Inc, 1942
71 WILSON, M G. AND EDWARDS, D J The vital capacity of the lungs and its relation to exercise *tolerance in children with heart disease Am J Dis Child*, 22: 443, 1921
72. WRIGHT, J The respir-aid bed in poliomyelitis Am J Nursing, 47: 454, 1947

Using the rabbit ear chamber technique, Ebert studied development of the local tubercle, the effects of tuberculin application to the tubercle and the development of serum sickness (16). Cortisone administration helped maintain better vascular tone, reduced damage to vascular endothelium, and reduced sticking of leukocytes to the endothelium. Subsequent leukocytic migration was diminished and a reduction in local exudate occurred. This was observed in all the above-mentioned groups. If, however, the inflammatory stimulus was intense enough, then no "cortisone effect" was noted on the process. Many studies along these lines have demonstrated that the administration of cortisone and ACTH to a variety of experimental animals reduces the total inflammatory exudate, regardless of the irritant, both in duration and extent. Thus, this exudate, containing among other things phagocytes, is less capable of handling the invader. It is not clear how these changes are brought about, but it does appear that this suppression of inflammation is nonspecific and does not necessarily represent an effect on antigen-antibody reaction Clark et al, studying the effects of cortisone, ACTH, and testosterone on rheumatoid arthritis, concluded that this antiphlogistic action in man was independent of the protein and electrolyte changes induced by these agents since the anti-inflammatory effects of cortisone and ACTH were not modified by testosterone administration as were the protein and electrolyte changes (7).

The effect, if any, of cortisone and ACTH on the phagocytic power of cells concerned with this important feature of bodily defense against infection is of importance. Here the data at hand are conflicting. The studies of Gordon and Katsch demonstrated a diminished uptake of thorium in the spleen of the adrenalectomized rat, with restoration of uptake by the administration of adrenal hormones (22). The administration of adrenal cortical extract, but not of desoxycorticosterone acetate, was accompanied by an increase in macrophages in the liver and in the spleen The amounts of cortical hormone employed were not comparable to the doses of cortisone used in subsequent studies to be mentioned. Using the *in vitro* test of phagocytosis of pneumococci by polymorphonuclear leukocytes from defibrinated human blood, Crepea and co-workers showed that in 9 out of 10 instances phagocytic activity was decreased in blood from patients who had received cortisone for a variety of diseases (9). Controlled amounts of type-specific antibody were added to each preparation Although the observed depression of phagocytosis was not of a high order, it was consistent Similar studies in our laboratory using a strain of *S aureus* have failed to demonstrate such changes (45) Moreover, cortisone and ACTH had no influence on the ability of mononuclear cells derived from oil induced peritoneal exudates of rats and rabbits to ingest oil particles (10). The phagocytosis of Beta hemolytic streptococcus injected into the pleural

gains access to the body, an inflammatory reaction is set up (14). This consists of margination and sticking of leukocytes to the wall of the capillaries in the invaded area, followed by their migration into involved tissues With them comes a *protein-rich edema fluid containing fibrin, antibodies,* and other substances. The process has potentialities of limiting the spread of the infection by containing it locally so that the body phagocytic cells, the polymorphonuclear leukocytes, and tissue and blood macrophages can ingest and destroy the invaders. Antibody, at times present in the edema fluid, may not be essential for recovery in the particular case, especially when antibiotics put less demand on the body for antibodies to control the infection In later stages of acute infections, and particularly in chronic infections such as tuberculosis, a wall of granulation tissue and, then, of fibrous tissue is thrown around the site. It can be reasoned teleologically that this is an attempt to limit spread of the infection through the body. Thus, any agent or situation that might intensify this inflammatory reaction will, in general, aid the body in localization of the process, and those that diminish the intensity of the reaction will favor dissemination to distant areas.

The ability of cortisone and ACTH to diminish this acute inflammatory reaction has been demonstrated by many techniques The beneficial clinical results achieved in patients with rheumatoid arthritis, rheumatic fever, asthma, and other diseases considered to be in some fashion the result of antigen-antibody reactions, led some to conclude that the effect was on diminishing "allergic inflammation" The writer thinks, as do others, that the lessening of inflammation is nonspecific and without regard to the incitant The evidence for this may be outlined as follows. The administration of cortisone to rabbits whose skin has been scarified, then rubbed with the violent irritant, croton oil, delays the appearance of the previously described elements of the acute inflammatory reaction (48) Margination and migration of leukocytes, capillary engorgement, and the appearance of edema fluid are delayed and diminished in intensity Effects of other stimuli, such as burns or turpentine, are similarly influenced by cortisone and ACTH (71) Tissue spreads taken from the site of the application of serum to sensitized rabbits demonstrate the paucity and delay in the appearance of the polymorphonuclear leukocytes and of other inflammatory cells (12) Woods and Wood, in a carefully controlled series of experiments, demonstrated that, in addition to blocking the inflammatory reaction in the eye resulting from protein anaphylaxis, or from bacterial hypersensitivity, cortisone and ACTH could also block with equal force the inflammation produced by the irritants glycerin and jequirity This action of cortisone can be invoked by local application as well as by systemic administration (82).

inflammation participates in the body's defense against micro-organisms, harm should result. Such, indeed, seems to be the case.

Effects on Bacteria

If an agent suppresses or enhances an infectious process, any effect on the specific micro-organism in question should be known. ACTH is obviously out of this part of our discussion, since its definitive action is mediated through secretions of the adrenal cortex. Cummings has shown that as much as 2.5 mg. per ml. of cortisone were required to inhibit the growth of virulent human tubercle bacilli (H37Rv) *in vitro;* whereas, only 0.25 mg per ml were needed to inhibit H37Ra, an avirulent varient (11). He suggested that this inhibition by large doses is probably due to the suspending agents and the 1.5 per cent benzyl alcohol used as a preservative. Comparable findings were noted by studying the respiration of the tubercle bacillus in the Warburg apparatus. In no concentration studied was there *growth stimulation of the organism. No effect of cortisone on the growth* of the hemolytic streptococcus was detected by Glaser (22), and we have been able to show no effect upon a variety of enteric and pyogenic organisms (45). Thus, any effects of cortisone and ACTH on an infection do not appear to be the result of direct action on the bacterial cell itself.

Effects on Immunologic Phenomena

Intensive investigations of the effects of cortisone and ACTH on certain immunologic phenomena have been recorded As with many of the experimental infections, conflicts result from studies on different species and from the use of different methods Mirick could detect in man no effect of ACTH on the antibody response to immunization with pneumococcal polysaccharide (49). In a beautiful series of experiments Germuth et al. showed that cortisone markedly inhibited the development of anaphylactic hypersensitivity of the Arthus type in rabbits receiving injections of crystalline egg albumin (21) The effects of ACTH were not of the same order. Circulating antibody was decreased, but the passive Arthus reaction was not interfered with. Moreover, these workers as well as others have demonstrated no effect on passively administered antibody (17). This suggests that the observed effects represent inhibition of antibody production rather than a destruction of that already existing Fischel found no alteration in susceptibility of guinea pigs to anaphylactic shock when treated with ACTH. He felt that the union of antigen and antibody was not altered by cortisone or ACTH (17) The development of the hemorrhagic necrosis of the Schwartzman reaction has been suppressed by ACTH. It did not, however, prevent development of the "state of reactivity" (68). The exact bearing these observations have on the topic under discussion remains in

cavity of rabbits appeared to be uninfluenced by cortisone administration (51).

Spain, Molomut and Haber administered 1 mg. of cortisone twice daily to mice for 5 days (70). On the third and fourth days, India ink was injected intraperitoneally, and the animals killed on the fifth day. The carbon particles were spread diffusely over the peritoneal cavity and visceral surfaces of the treated animals with only occasional foci on the surfaces of liver and spleen. The particles in the untreated animals were in omental, mesenteric, and retroperitoneal regions, and the superior mediastinal nodes were laden with the carbon. They attributed these findings to diminished quantities or function of phagocytes in the treated groups as well as to the possible antihyaluronidase effect of cortisone preventing diffusion of the particles.

Lurie et al found that the intravenous administration of carbon to rabbits that had received cortisone for 34 days resulted in an increased amount of carbon by weight in the spleens and livers (44). They attributed this to enhancement of phagocytosis by cortisone. From the experiments, such a conclusion is not permissible Histologic studies of lymph nodes in experimental hemolytic streptococcal pneumonia in rats demonstrated a delay in the appearance of the polymorphonuclear leukocytic exudate, but when it did appear phagocytosis did not seem to be altered (22) It seems reasonable to state that with the evidence at hand no consistent effect on phagocytosis is exerted by cortisone and ACTH.

Hyaluronidase, or the spreading factor of Duran-Reynals, is present in certain bacteria Whether it plays a significant role in the spread of bacterial infections is not established Cortisone has been shown to inhibit the spreading properties of hyaluronidase as measured by diffusion of particulate matter through the skin, or by its appearance to the site of inflammation (4) 24 to 48 hours of cortisone therapy before intravenous administration of hyaluronidase was necessary to invoke the inhibitory effect. This suggested to Benditt and collaborators that the cortisone effect is on mesenchymal cells which, in turn, alter intracellular substance.

Adequate studies have demonstrated the inhibitory effects of cortisone and ACTH on the appearance and development of granulation tissue. In instances where this fibrous reaction is an intimate part of the evolution of an infectious process, its inhibition or alteration by the administration of cortisone or ACTH might alter the outcome of such infection

"y suppress
iving such
ions where
n of these
where the

extensive inflammatory lesions in the myocardium and kidneys, with dense deposits of cocci in both organs.

Pneumococcal Infections

Kass and co-workers demonstrated that cortisone in doses of 5 mg. a day and ACTH of 2.5 mg. twice a day had no profound effect on the outcome of pneumococcal infection in white mice. It was their impression that those animals that received ACTH tended to live a little longer and those treated with cortisone to die sooner and to have a slightly higher group mortality than did the controls (32). It is to be remembered that pneumococcal infection in mice is particularly virulent with a rapid and exceedingly high mortality. Partial protection of the mice with type specific anti-pneumococcal serum was then studied Infection of these animals with virulent pneumococci and treatment with cortisone or ACTH afforded no protection There was a higher mortality in the cortisone treated group than in the ACTH treated group In another study, it was demonstrated that 0 5 ml. of aqueous adrenal extract given to mice with pneumococcal infection and treated with subcurative doses of sulfadiazine resulted in slightly longer survival times, but had no effect on ultimate mortality. This was interpreted as supplying an adequate amount of adrenal hormones to correct the partial adrenal insufficiency presumed to have resulted from the infection (77) The results of cortisone administration to rats infected endobronchially with pneumococci were the same in type and magnitude as described for the hemolytic streptococcus by Glaser et al. (22).

Tuberculosis

The alteration of tuberculosis in the experimental animal by ACTH and cortisone has been studied more intensively than has any other infection Some of the conflicting observations must be examined in the light of extreme species variation in susceptibility to strains of the tubercle bacillus in these animals

Cortisone administered for 40 days was responsible for a disseminated tuberculosis with a high mortality in albino rats infected with the H37Rv strain By way of contrast, the control rats infected with the same organism developed a benign disease with localized granulomata in various organs In the cortisone treated group the lesions were more diffuse, contained many more tubercle bacilli, fewer inflammatory cells and less fibroblastic proliferation than those of the untreated controls (47) Streptomycin therapy protected the cortisone treated rats with survival rate and histologic picture comparable to those of rats infected but not given cortisone (11). There were no deaths in the animals uninfected, but given cortisone for comparable periods of time No pyogenic lesions resulting from secondary

question. Particularly, one doubts that the effects on antibodies have a role in the observed clinical and experimental results with ACTH and cortisone in infections since it is probable that antibody could have little influence on the acute experiments to be outlined.

EFFECTS ON VARIOUS SPECIFIC INFECTIONS IN EXPERIMENTAL ANIMALS

Cortisone and/or ACTH have been administered to many different experimental animals with a variety of induced infections. A perusal of the results will facilitate understanding of the findings in man and will, to a certain degree, permit prediction of future observations.

Bacterial Infections

Mice infected intranasally with Group A hemolytic streptococci and treated with 2 5 mg. of cortisone daily had significantly shorter survival times than did the untreated controls (22). This was noted when cortisone was begun the day of infection or five days before infection. All animals in both groups, however, died. Again, a less favorable survival time was found in white rats with streptococcal pneumonia and treated with cortisone. All the treated animals had succumbed by the time the mortality rate was only 50 per cent in the control group. The lesions in the cortisone treated group were more hemorrhagic and pleural infusions were frequently bloody. Histologically, the edema fluid was more abundant, the cellular exudate less, and the number of streptococci in alveoli markedly increased The differences were particularly striking 18 to 24 hours following inoculation Studies of the peripheral blood count of rats demonstrated a leukopenic effect of cortisone There was an abundance of polymorphonuclear leukocytes, however, suggesting to the authors that their egress from the blood stream into the lesion was in some way impeded by cortisone. When small inocula of streptococci were used, bacteremia occurred much earlier in the cortisone treated animals than in the controls in which it was a manifestation of the terminal event (22).

Mogabgab and Thomas observed that the administration of cortisone in doses of 50 mg a day to rabbits injected intradermally with Group A hemolytic streptococci resulted in a uniformly fatal infection contrasted with an innocuous course in the controls (50)

Further studies by Thomas and his group are of interest In rabbits immunized with streptococci prior to infection and cortisone treatment, bacteremia was slight and transient and all animals survived, indicating that the previously described enhancement of infection was not brought about by interfering with the immune defenses These animals did develop

tuberculin reaction in man and in animals has been noted on many occasions (25, 66). There are two schools of thought as to the modes of action in such tuberculin suppression. There are those who believe that it represents interference with the antigen antibody reaction; others favor the hypothesis that it reflects the ability of these hormones to diminish the intensity of an inflammatory reaction, regardless of the stimulus. At the present time, no completely adequate explanations are at hand for the deleterious effects in tuberculosis in animals.

Miscellaneous Infections

Guinea pigs were given 0.1 ml of a 24-hour culture of coagulase positive *S. aureus* in each of two sites. Similar injections were given on 5 consecutive days. The cortisone treated group had received 5 mg. of the drug a day for 30 days preceding inoculation. In this treated group, the lesions were more severe, induration greater, and healing delayed (40). Abernathy studied the effects of cortisone on infections with *Brucella abortus*, *suis*, or *melitensis* in mice, guinea pigs, and rabbits Use of this hormone resulted in increased death rate and a wider distribution of lesions throughout the body. The hepatic granulomata became necrotic. No effect on the agglutinin titers was demonstrable (1)

Friedlander isolated strains of alpha hemolytic streptococci which produce purulent arthritis in mice following intravenous injection. Cortisone in doses from 0 025 to 0 015 gm. per kg. for 5 days, beginning the day before infection, produced statistically significant increases in the incidence of arthritis and in mortality. Doses below this were without significant effect (20). Similar findings are recorded by Kuzell and Mankle in arthritis produced by pleuropneumonia-like organisms in rats (41).

The lesions of experimental syphilis in the rabbit were strikingly altered by the administration of cortisone (76). In treated animals, the dermal syphilomas became smaller within 48 hours, then paler, soft and spongy, and filled with a mucinous material tentatively identified as hyaluronic acid The lesion then enlarged; and, after 15 to 25 days of cortisone medication, ruptured, extruding the material. Darkfield examination of this revealed *T pallida* in larger numbers than in untreated controls. Upon biopsy, a decrease in inflammatory cells was noted in the cortisone treated animals. Although the Wassermann titer was significantly lower in the treated animals, the treponema immobilizing antibody showed no difference in titer Upon withdrawing cortisone, there was a rebound phenomenon with lesions becoming larger than those of controls at comparable stages of infection Turner cautions against such reactions in humans given cortisone. He concludes that cortisone partially suppresses the response of the host while the treponemes multiply in an unrestrained manner (76).

invaders were observed when animals were sacrificed. This would tend to exclude the possibility in these experiments that the increased susceptibility to secondary infection in cortisone treated tuberculous animals might be responsible for the higher mortality. Spain and Molomut were able to demonstrate more extensive disease in guinea pigs infected with tubercle bacilli and treated with cortisone than in those infected but not given cortisone (69). In contradistinction to the previously mentioned rat experiments, streptomycin did not protect guinea pigs against the deleterious effects of cortisone. Perhaps this discrepancy can be explained by the fact that the albino rat is naturally resistant to tuberculosis while the guinea pig is highly susceptible. Bloch and his collaborators demonstrated more extensive tuberculosis in cortisone treated guinea pigs than in controls (5). The studies of Lemaistre and Tompsett are contradictory. They were unable to demonstrate any enhancing action of cortisone or ACTH upon the initiation and early progression of a severe tuberculous infection in guinea pigs (42) These variable results in guinea pigs may be attributable to the fact that tuberculous infection in the guinea pig is such an exceedingly virulent one that degrees of enhancement would be difficult to assess

Investigations using other animals show the trend toward worsening of the disease Cortisone given to mice infected with H37Rv changed a nonprogressive, predominantly proliferative disease into a usually fatal disease characterized by large, coalescing, necrotic lesions swarming with tubercle bacilli. Similarly, cortisone given after a large inoculum of tubercle bacilli which produces an acute fatal infection in mice caused death earlier than in the controls (26) Lurie et al studied the actions of cortisone in a strain of rabbits of a high genetic susceptibility to tuberculosis. The animals were infected by the inhalation route (44) In animals treated with cortisone, many more pulmonary tubercles and more caseation were noted than in the untreated controls Moreover, there were many more tubercle bacilli in the lesions The tubercles were smaller in size and showed less spread to the regional nodes than in the controls The lesions noted were, in their experience, similar to those in the genetically resistant strain. However, one finds it difficult to reconcile more tubercles and a larger number of *tubercle bacilli with resistance to the infection.* The discrepancy in recorded results can be attributed to difference in species and in their response to infection. The weight of evidence indicates, however, that cortisone and ACTH definitely enhance tuberculous infections in most experimental animals Inhibitory effects of cortisone or of ACTH on the systemic response of sensitized animals to tuberculin have been noted in many animals *This* was demonstrated for classical tuberculin shock or for the pulmonary changes following intratracheal instillation of tuberculin into sensitized rabbits (11, 60) Moreover, reduction of the intensity of the intracutaneous

were no changes in the glucose tolerance although other signs of adrenal hyperfunction appeared (2).

Thygeson, Geller, and Schwartz studied the course of herpes simplex keratitis in the rabbit. Cortisone instilled locally into the eye, or given systemically, provoked either no change or a lesion of greater intensity with more rapid onset of fatal encephalitis (75).

Experimental Parasitic Diseases

Of interest are the effects of cortisone on certain experimental parasitic diseases. Schmidt and Squires, in studying the course of *P. cynomolgi* malaria in the rhesus monkey, found that cortisone had no effect on the initial degree of parasitemia in the primary attack (63) However, in the "post crisis" phase, beginning approximately 16 to 20 days after infection when parasites were barely detectable in control animals, the cortisone group showed continued heavy parasitemia with a slow, irregular decline for a period of 12 to 16 days In control animals, the post crisis period of relatively insignificant parasitemia lasting 10 to 14 days was followed by secondary rises not at all comparable to those of cortisone treated animals. Cortisone given to monkeys with latent infections initiated severe recrudescences No detectable difference could be noted in the morphology of the parasite or in its asexual cycle The picture recorded in the cortisone group was comparable to that seen in splenectomized, infected monkeys The authors thought that the exhaustion of lymphoid elements with its subsequent marked diminution in production of macrophages was largely responsible for the observed results. This explanation cannot be completely accepted since it is not certain that macrophages are derived from lymphoid elements No apparent effect on phagocytosis of the parasites was observed The absence of any effect on the initial stages from beginning parasitemia to crisis, they attribute to the lack of effect of cortisone on "innate immunity" Redmond has described comparable findings in pigeons infected with *P relictum* In controls, parasitemia reached a peak within 8 days with recovery of the birds, whereas, in cortisone treated birds, the parasitemia persisted and within 3 weeks over 50 per cent were dead (58)

There are conflicting results in experimental trypanosome infection. Cortisone had no effect on the fatal outcome, nor on the terminal hypoglycemia, of *T equiperdum* infection in the fasting rat (73) On the other hand, cortisone increased mortality and intensified parasitemia of *T. cruzi* infection in dba mice Pentaquine did not counteract the deleterious effects of cortisone (27)

A latent *T vickersae* infection in rhesus monkeys was unmasked by doses of cortisone as small as 5 mg a week for 6 weeks Encephalomyelitis, myo-

Viral Diseases

It is impossible to determine whether or not cortisone has a direct effect on virus multiplication in view of the necessity of using cells for their cultivation *in vitro*. In the chick embryo Kilbourne and Horsfall have demonstrated an increased titer of mumps and influenza A and B viruses in eggs injected with cortisone (35). Such action does not necessarily indicate stimulation of viral growth, and could be explained on the basis of some less well-understood action of cortisone on the embryonic tissues.

Kass and his co-workers detected no effect of cortisone and ACTH on influenza virus infection in mice, nor on the multiplication of virus in the lungs (32). Kalter et al showed a lower titer of virus in the lungs of mice infected with influenza and treated with cortisone for 7 days or ACTH for 10 days previously (28) This they attributed to an anti-anabolic effect of these hormones in view of the fact that the administration of testosterone, which increases protein anabolism, increased virus multiplication; whereas castration decreased it. These observations are of considerable interest for they show how the alteration of one of the metabolic processes in the body can affect virus multiplication, presumably by altering the metabolism of the cell in which the virus grows

More striking effects have been demonstrated in other experimental viral infections By giving a single injection of 2 to 5 mg. of cortisone 2 days before inoculation, Kilbourne and Horsfall were able to produce a lethal infection with Coxsackie virus in adult mice (36). Heretofore, only suckling mice had proved susceptible to Coxsackie infection. With this technique, however, it should be much more practical to attempt further Coxsackie isolation Indeed, these workers were able to recover the virus directly from human feces using adult mice pretreated with cortisone. Here, then, is a practical laboratory method contributed by use of cortisone.

Using adrenal cortical extract which can be compared only roughly with cortisone, Vollmer and Hurlbet showed that the extract in amount equivalent to 250 micrograms of cortisone a day increased by 50 per cent the mortality of mice infected subcutaneously with Japanese B encephalitis virus. No effect was noted when the virus was injected intraperitoneally, nor when the dose of cortical extract was lower (78)

More conclusive results are at hand concerning the influence of cortisone and ACTH on the outcome of poliomyelitis infections A single injection of cortisone accelerated the infection with the MEF 1 strain of poliomyelitis virus in mice and increased susceptibility in hamsters A violent and uniformly fatal infection was produced ACTH did not produce such changes (64). On the other hand, Ainslee, Francis, and Brown found that ACTH slightly accelerated the onset of paralysis in monkeys and in mice There

Bacterial Infections

Kass and his collaborators have described the effects of ACTH on the course of *pneumococcal pneumonia* (33). In 3 patients, there was a prompt drop in fever, sweating not unlike that of a crisis, and striking subjective improvement. When the patients were afebrile, pneumococci were abundant in the sputum, with little if any evidence of phagocytosis. In 1 patient, there was bacteremia 12 and 36 hours after he was clinically well although there was a negative blood culture before ACTH was administered This study is an interesting one which cannot be entirely explained. Perhaps the ACTH suppressed all of the usual signs of infection, suppressed the intense inflammation, and allowed bacteremia to develop. It is of interest that the development of agglutinins was in no way interfered with. One patient with Type VII pneumococcal pneumonia, given cortisone by nebulization in amounts up to 8 mg. every 30 minutes, became afebrile after 12 hours (59). The treatment was continued for 33 hours. Blood culture was positive at 12 hours and negative thereafter. X-rays showed progression during the first 12 hours of treatment with clearing beginning thereafter. When the amount of cortisone was decreased to 5 mg. at each inhalation, the temperature began to rise and the patient worsened clinically. He had a good response to aureomycin.

Thus, although amelioration of clinical symptoms may ensue from cortisone treatment of pneumonia, a cure does not take place. Indeed, it must be emphasized that bacteremia may *develop* although the patient is afebrile and feeling well. The dangers of this are obvious.

Cortisone was given in conjunction with chloramphenicol to 8 patients with *typhoid fever* by Smadel, Ley, and Diercks (67) This combination resulted in a more prompt amelioration of the acute symptoms than did the administration of the antibiotic alone. Those receiving 300 mg of cortisone the first day, followed by 100 mg a day for 4 days, became afebrile within an average of 15 5 hours, while those receiving smaller doses on the first day required an average of 50 2 hours for the same effect. There were no perforations, but 1 patient had intestinal hemorrhage. There were two relapses, more than were expected from a comparable group treated with chloramphenicol alone In a companion study, Woodward and his collaborators administered cortisone alone to 7 patients with typhoid fever (83). In 6 patients, there was defervescence, requiring from 1 to 100 hours. In the seventh there was no effect The positive blood cultures became negative by the fifth day of therapy One relapse occurred and was promptly controlled by another course of the hormone The stool cultures required as long as 26 days to revert to negative. Roche gave 80 mg. of ACTH a day to 2 patients with typhoid and 1 with paratyphoid fever (61) In one with typhoid, the temperature fell to normal within 24 hours with occasional

carditis, and less often myositis, neuritis, and lymphadenitis, were observed. The monkeys receiving no cortisone showed no trypanosomal lesions (81). This is an example of a latent parasitic infestation activated by cortisone. Other latent infections in various animals have been activated by cortisone or ACTH (51). These findings indicate the pitfalls that one might encounter in animal experiments making some results difficult of interpretation. They also demonstrate the delicate host-parasite symbiotic relationship that can be upset by these agents.

CLINICAL APPLICATIONS

Cortisone and ACTH have marked potentialities for affecting the outcome of an infectious process. The results to date can be examined conveniently under the categories of: 1) Specific treatment of infectious diseases; 2) Effects on "latent infections", and 3) Properties of suppressing signs and symptoms of infection.

Cortisone and ACTH Effects Capable of Masking Signs of Infection

The clinical recognition of an infectious process is facilitated by the appearance of signs and symptoms attributable to the reaction of the host to the disease. An example is the sense of being "unwell", a usual accompaniment of any infectious process, especially an acute one. Cortisone and ACTH have a capacity to produce a striking improvement in the sense of well-being, irrespective of the disease being treated. This phenomenon occurs within a few hours after the first injection of either agent. This increased sense of well-being is often the first change during cortisone treatment of patients with rheumatoid arthritis. One can see the confusion such nonspecific amelioration of symptoms could cause in a patient with an infection who should appear ill. The implications of this will be noted later

Kass and Finland demonstrated an antipyretic effect in man of ACTH on fever induced by the intravenous administration of typhoid vaccine (29) Although fever was not completely abolished, it was significantly reduced and the anticipated chill did not occur. They demonstrated an antipyretic effect of ACTH in rabbits receiving either typhoid vaccine or influenza virus intravenously The studies of Duffy and Morgan, using 15 mg of ACTH and 5 mg. of cortisone a day for 2 days, showed significant lowering of the febrile response of rabbits to shigella endotoxin However, using 5 mg. of ACTH or 5 mg. of cortisone for 24 hours before injection, they found the febrile response to be aggravated There are no plausible explanations for these discrepancies (15). Cortisone or ACTH often abolishes febrile response evoked by a variety of illnesses in man, only for it to return upon discontinuation of medication (30, 46) The masking of fever makes recognition of an underlying infection difficult with resulting treacherous situations of great clinical hazard.

while in the other extension was questionable Lemaistre et al. studied 7 patients with far-advanced pulmonary tuberculosis (42). Four of them had tuberculous laryngitis and another tuberculous empyema. Cortisone was given to 3 and ACTH to 4, in doses of 100 mg daily for an initial 10 day period. In all instances, there was defervescence, return of appetite, and an increased sense of well-being These disappeared upon discontinuing the drugs. The laryngeal lesions showed decrease in edema and exudate with evidence of epithelialization. The pulmonary lesions showed slight increase in translucency. However, when the hormones were discontinued, the disease reverted to its original status. Indeed, in some instances it was more severe. There was temporary suppression of the tuberculin reaction in 3 patients. This may represent only suppression of inflammation rather than true suppression of hypersensitivity, although the effect was noted in two instances for 3 to 4 weeks after cessation of the hormone. Kinsell and co-workers administered cortisone or ACTH in combination with streptomycin to several patients with tuberculous meningitis (38). It was their impression that there was a diminution in "morbidity and mortality". They also thought that some patients with extensive tuberculosis might be prepared for surgical procedures by the combined hormone-chemotherapy regimen, although small doses of the hormones were recommended No harmful effects on this regimen were noted on genito-urinary tuberculosis. Further details from Kinsell's group are to be recorded at an early date and will be watched for with considerable interest.

I have heard of several other patients receiving either cortisone or ACTH plus streptomycin in an attempt to prevent formation of the inflammatory and fibrous barrier around the tuberculous focus and permit better access of the chemotherapeutic agents. There are no results that warrant analysis at this time, since these studies are in the early experimental stages only. The deleterious effects of these substances on tuberculosis is unquestioned; whether they can be obviated by chemotherapy remains to be seen—it is doubted

It has long been thought that the Waterhouse-Friedrichsen syndrome seen in fulminating meningococcemia was caused by acute adrenal insufficiency resulting from bilateral adrenal hemorrhage with subsequent tissue destruction Such an idea is no longer tenable in view of the many reported recoveries. If such complete adrenal destruction had existed, the patients should have developed Addison's disease They have not. Furthermore, some patients dying with this syndrome have not had widespread adrenal hemorrhage However, degenerative changes have been demonstrated in the adrenal cortex. Dramatic recovery from the Waterhouse-Friedrichsen syndrome by 2 patients who received cortisone in addition to the other therapies indicates that such therapy may have a role in over-

secondary rises to 38°C. for 1 week after the drug was stopped. Blood cultures remained sterile The other case of typhoid showed little effect. The case of paratyphoid fever responded in a not too dramatic fashion and had persistently positive blood cultures.

How can the apparent beneficial effect in some cases be explained? Since *S typhi* is an intracellular parasite, one might postulate that cortisone exerts its antianabolic effect on the host cells, comparable to that recorded for the influenza virus, and that such cellular protein deprivation makes for a less favorable environment for the microorganism Such is purely conjecture Whatever the explanation may be, ACTH or cortisone must not be used alone in the treatment of typhoid fever. Moreover, one doubts the wisdom of its use in conjunction with chloramphenicol, particularly since perforation at the widespread intestinal ulcerations could easily be provoked and masked by cortisone. In a patient gravely ill with typhoid fever, however, one might be justified in giving a single injection of 300 mg. of cortisone to control the acute "toxicity" of the disease until the antibiotic could take effect Although favorable results have been recorded in patients with ulcerative colitis treated with cortisone or ACTH, this author feels a word of caution is indicated Perforations of the friable colon have occurred and signs of the peritonitis have been masked by cortisone (45). Actually, roentgenograms of the colon have demonstrated spread of lesions while the patient was receiving cortisone and had become asymptomatic (65)

An epidemic of exudative streptococcal pharyngitis was studied by members of the Streptococcal Disease Laboratory at Fort Warren, Wyoming (24). 87 cases served as controls and 87 were given cortisone The first 17 received a total of 500 mg in 50 mg doses The remaining received 600 mg. in 100 mgm doses There was no difference in symptoms or signs of infection in the two groups and those receiving cortisone had fever for a longer period. Of the suppurative complications, 1 occurred in the treated group and 3 in the controls. Rheumatic fever developed in 2 of the treated and in 5 of the controls They did not consider this to be of significance The immunological response, as measured by antistreptolysin titer, was not appreciably altered by the doses employed They suggest that significant depression of antibody might have reduced the incidence of rheumatic fever

The results of treatment of proved tuberculosis are of considerable importance. Freeman and his associates administered ACTH to 2 patients with pulmonary tuberculosis in doses of 100 mg a day for 3 weeks, 75 mg a day for 1 week, then 100 mg a day for 5 weeks (19) In both, fever disappeared, productive cough diminished and appetite improved The sputum remained positive for tubercle bacilli in both instances. In 1 patient, there was definite extension of the pulmonary lesion during ACTH therapy,

toms returned when the drug was discontinued, then remitted when it was exhibited for the second time. There was no effect on the eosinophilia (62). The administration of ACTH to 3 patients with malaria inoculata resulted in an intensification of parasitemia with a slight decrease in fever (31). No accounts have appeared of the treatment of amebiasis, and this is good These substances should not be given to anyone with amebiasis in view of the multiple bowel ulcers which could easily perforate under the influence of cortical hormones.

In 1 patient with Rocky Mountain spotted fever given 300 mg. of cortisone on the fifth day of disease and 150 mg. a day for 2 days thereafter, there was only slight temperature lowering effect and no effect on rash or headache. When terramycin was administered, there was a prompt response (57). In a series of 9 other cases, cortisone was given in addition to chloramphenicol in an attempt to reduce the "toxemia" of the disease (84). Cortisone was given in an initial injection of 200 mg. followed by 2 100-mg. doses at 6-hour intervals. The total amount did not exceed 400 mg. There was a striking alleviation of symptoms and toxicity with a return of the temperature to normal in an average of 1 7 days The comparable time for patients treated with chloramphenicol alone to become afebrile ranged from 2.5 to 4 days. The authors rightly conclude that cortisone should not be used routinely. In cases seen late in the course of the disease when "toxemia" is such a problem, cortisone or ACTH might be useful adjuncts. The dangers of the compounds must be carefully weighed against the possible therapeutic effect Many more cases must be so treated before effect on mortality is evaluated.

Cortisone and ACTH Masking and Enhancing an Infection

It appears that cortisone and ACTH not only can incite the development of an infection, but can enhance its progress and make its recognition difficult by their suppression of some of its cardinal signs and symptoms There are many well-documented instances of such a train of events. We have observed the development of pneumococcal pneumonia in the terminal stages of disease in a patient treated with cortisone for lymphoma and in one for multiple myeloma (80). In each, the signs and symptoms of pneumonia were lacking and the febrile response was suppressed. One patient receiving cortisone for burns died of an overwhelming *Ps aeruginosa* bacteremia while still afebrile. Insignificant furuncles have been noted to spread into diffuse cellulitis under hormonal therapy, healing only when medication was discontinued. Patients have died with unrecognized infections, such as pericarditis, staphylococcic sepsis, pneumococcic sepsis, empyema, and peritonitis (34, 46, 13) Absence of fever and local signs of the infections masked the processes The development and spread of

coming temporary acute adrenal insufficiency and "toxemia" (53, 54). However, similar dramatic recoveries have occurred with chemotherapy and electrolyte replacement alone The cortisone supplement is still in the experimental stage. Further use in patients with such fulminating infections should be explored with caution. Cortisone had no effect on the mortality of mice infected with meningococci, but it did diminish adrenal congestion (52). Thomas and Good have produced bilateral cortical necrosis of the kidneys in rabbits treated with cortisone and given meningococcal "toxin" intravenously (74).

Boling et al have given ACTH or cortisone in conjunction with appropriate chemotherapy to 6 patients with peritonitis (6). In all, there was an immediate reduction or disappearance of signs of systemic toxicity. In 1 child with peritonitis following appendicitis, ACTH reduced fever, toxicity, and abdominal rigidity, and permitted operation within 48 hours. There was no inflammatory reaction despite an abdominal cavity filled with purulent material. One wonders whether such a host reaction is beneficial. Teleological reasoning would suggest not. These authors do emphasize the ability of these hormones to mask advanced and advancing infection, and urge extreme caution in their use in such instances

Viral Diseases

Thirty-five of a group of 70 patients in the acute stage of poliomyelitis were treated with ACTH while the remainder received a placebo There was no effect of the hormone as measured by temperature response, development of paralysis, progression, or early residuals (8). We have administered cortisone to 7 patients with mumps orchitis with the idea of combatting some of the inflammatory edema and of bringing about a more rapid symptomatic recovery (45). It is the clinical impression—usually fraught with errors—that, though the series is small and without controls, the fever and pain disappeared more rapidly (24 hours) than was to be expected. A smallpox epidemic in Holland afforded Stolte and Sas the opportunity to study the effects of ACTH in combination with chloramphenicol on 3 patients (72). The inflammation around the lesions was definitely less than in controls given chloramphenicol alone, but there was no detectable difference in the pox They concluded that its use was fraught with danger in this disease

Patients treated with ACTH for viral pneumonia had a rapid fall in temperature and a return of the sense of well-being However, it was not thought that the medication altered appreciably the natural course of the disease. Symptoms returned when ACTH was stopped (33)

A patient with *trichiniasis* treated with cortisone had a striking fall in temperature and amelioration of symptoms within 4 days The symp-

obtained from the sputum. Two infections were probably precipitated by the hormones in question—Friedlander pneumonia and pulmonary tuberculosis. Many such cases have been alluded to in the literature or have come to my attention by word of mouth. The weight of evidence indicates that cortisone and ACTH may reactivate quiescent or latent tuberculosis. Therefore, it appears that these hormones should not be given to patients with a known history of tuberculosis. Furthermore, careful x-ray studies should be made in cases where prolonged drug administration is contemplated.

A number of other bacterial complications are being noted. These have occurred with either cortisone or ACTH, used for burns, collagen diseases, and a variety of other entities. In the majority of instances, doses in the range of 100 mg. a day have been used and for at least a week. There are no figures on the frequencies of such complications. In our experience, they occur more often than do hypokalemia and psychosis. How then can they 1) be avoided, and 2) be recognized? Certainly, if the indication for cortisone or ACTH is in doubt, it is best to err on the conservative side and to avoid administration. Moreover, if there is chronic pulmonary suppuration or "quiescent" tuberculosis, extreme caution must be exercised in any hormonal therapy. A careful check on the clinical progress along with frequent chest films should be a part of the routine in any patient receiving either of these hormones. A negative tuberculin reaction at the start of therapy would be comforting to the physician. During therapy, the first indication of a suppurative or tuberculous complication should call for immediate discontinuation of medication and for the institution of appropriate anti-infective measures But above all, the physician must be on guard for masked infections that develop and progress with few, if any, classic signs and symptoms to aid in their recognition. It has been said somewhat facetiously, but with a modicum of truth, that these substances "permit the patient to walk to the morgue".

Are There Differences between Cortisone and ACTH?

Many of the clinical results that have been achieved with these two substances are quite similar It is apparent that cortisone does not represent the true cortical "S" hormone and that ACTH stimulates the adrenal to secrete things other than the "S" hormone. Considerable confusion exists as to whether the effects of the two agents are the same in infectious processes Much of the conflicting information stems from the fact that animals of various species respond differently to these substances. As an example, much larger doses of ACTH than cortisone must be used to produce an eosinopenic effect in mice In this writer's opinion, one should consider that for practical purposes the end result in human infections is the same regard-

decubitus ulcers in subjects receiving ACTH or cortisone warrant special attention since healing is markedly delayed and infection permitted to spread further (3). Peptic ulcers have perforated during cortisone and ACTH therapy with few signs to aid in recognition of the ensuing peritonitis. Patients with ulcerative colitis have perforated, soiling the peritoneum with feces, yet showing little that would help in recognition of peritonitis (45).

The effects of cortisone and ACTH on tuberculosis have been indicated. Of graver import is reactivation in patients receiving these compounds for unrelated diseases. A presumably quiescent tuberculosis rapidly disseminates. Among the first patients we treated with cortisone was one with periarteritis nodosa who, after 2 weeks of therapy, had hemoptysis with sputum positive for acid-fast bacilli and a rather rapid spread of tuberculosis. His chest films before hormone therapy showed a "healed" apical lesion. Another patient being treated for a lymphoma was found at autopsy to have miliary tuberculosis with widespread dissemination (80). Kleinschmidt and Johnston reported miliary tuberculosis at autopsy in a patient who received cortisone for 4 weeks for a "febrile illness with arthritic manifestations" (39). The cortisone had been discontinued for 3 months, and the age of the tubercles was estimated at about 8 weeks They indicated that absolute cause and effect relationship was lacking, but the facts indicated to them that cortisone precipitated the development of tuberculosis

In the September 15, 1951, issue of the Journal of the American Medical Association are three similar case reports In one, spread of a pulmonary lesion with ultimate death of the patient was noted. However, the patient had lower lobe disease, negative sputa examinations, and no autopsy confirmation was to be had (37) A patient studied by Popp, Ottosen, and Brasher had quiescent, fibrotic, pulmonary tuberculosis and rheumatoid arthritis (56). Over the 4-year period she had been observed, there had never been a positive sputum by smear, culture, or guinea pig inoculation She was given 2500 mg of cortisone for arthritis, the duration of treatment being 29 days. There was no change in her chest as determined fluoroscopically. Three weeks later, cortisone was recommenced After 1500 mg had been given over a period of 17 days, she had hemoptysis. X-ray now revealed cavity formation; and, for the first time, sputum smears were positive for acid-fast bacilli. Fred et al reported the case of a 58-year-old man who received both cortisone and ACTH for rheumatoid arthritis (18). His chest film was clear before therapy. After 21 days of ACTH, fever rose and he developed cough. Cortisone was then added to his regimen. A chest film taken 10 days later indicated extensive consolidation in the right upper lobe. *K. pneumoniae* were recovered from the sputum, but were eradicated with appropriate chemotherapy Shortly thereafter, tubercle bacilli were

14 DUBOS, R. J., Bacterial and Mycotic Infections in Man, p 61, 90 Philadelphia, Lippincott, 1948
15 DUFFY, B. J., JR AND MORGAN, H. R.: ACTH and cortisone aggravation or suppression of febrile response of rabbits to bacterial endotoxin Proc Soc. Exp Biol & Med, 78: 687, 1951.
16 EBERT, R H. *In vivo* observations on the effect of cortisone on experimental tuberculosis using the rabbit ear chamber technique Tr. Nat. Tub. Assn, 140-152, 1951.
17 FISCHEL, E E: The relationship of adrenal cortical activity to immune responses. Bull N. Y. Acad Med, 26: 255-260, 1950
18 FRED, L, LEVIN, M. H., RIVO, B. J. AND BARRET, T. F · Development of active pulmonary tuberculosis during ACTH and cortisone therapy. J. A. M. A, 147: 242-246, 1951.
19 FREEMAN, S, FERSHING, J., WANG, C. C. AND SMITH, L C · The effect of ACTH on patients with pulmonary tuberculosis Proc. First Clin. ACTH Conf, 509 Blakiston, 1950
20 FRIEDLANDER, H.: Effect of cortisone acetate on experimental purulent arthritis of white mice J. Inf Dis, 89: 26-30, 1951.
21. GERMUTH, F. G, JR, OYAMA, JIRO AND ATTINGER, B: The mechanism of action of 17-hydroxy-11-dehydrocorticosterone (Compound E) and of the adrenocorticotropic hormone in experimental hypersensitivity in rabbits J Exp Med, 94: 139-170, 1951.
22 GLASER, R. J, BERRY, J W, LOEB, L H AND WOOD, W. B, JR.: The effect of cortisone in streptococcal lymphadenitis and pneumonia J Lab. & Clin. Med, 38. 363-373, 1951
23 GORDON, A S AND KATSH, G F · The relation of the adrenal cortex to the structure and phagocytic activity of the macrophagic system Ann N Y Acad Sci, 52: 1-30, 1949
24 HAHN, E O, HOUSER, H B, RAMMELKAMP, C. H., JR, DENNY, F. W AND WANNAMAKER, L. W.. Effect of cortisone on acute streptococcal infections and poststreptococcal complications J Clin Inv., 30: 274-281, 1951
25 HARRIS, S AND HARRIS, T N.· Effect of cortisone on some reactions of hypersensitivity in laboratory animals Proc Soc Exp Biol & Med, 74: 186, 1950
26 HART, P. D AND REES, R J W Enhancing effect of cortisone on tuberculosis in the mouse Lancet, 2: 391, 1950
27 JARPA, A., AGOSIN, M, CHRISTEN, R AND ATIAS, A V Ensayo de quimoterapia de la enfermedad de chagas VII Cortisona y fosfata de pentaquina Biol de Inf Parasit Chil, 6: 23, 1951
28 KALTER, S S, SMOLIN, H J, MCELHANEY, J M AND TEPPERMAN, J Endocrines and their relation to influenza virus infection J Exp Med, 93: 529-538, 1951.
29 KASS, E H AND FINLAND, M Effect of ACTH on induced fever New Eng J Med, 243: 693-695, 1950
30 KASS, E H AND FINLAND, M The role of adrenal steroids in infection and immunity New Eng J Med, 244: 464-470, 1951
31 KASS, E. H, GEIMAN, Q M, INGBAR, S H, LEY, A B, HARRIS, J W. AND FINLAND, M Some diseases which may be activated by ACTH—Observations on sickle cell disease and malaria Proc Second Clin ACTH Conf. J Mote, Ed, 2: 376 Blakiston, 1951
32 KASS, E H, INGBAR, S H AND FINLAND, M Effects of adrenocorticotropic hormone in pneumonia· clinical, bacteriological, and serological studies Ann. Int. Med, 33: 1081-1098, 1950.

less of the hormone used. Infections can be precipitated, enhanced, or masked by either.

SUMMARY

Cortisone and ACTH have profound effects on a variety of experimental and clinical infectious processes. In the main, these are deleterious, but on occasion may be favorable by lessening "toxicity" in the host. The harmful effects, brought about by actions on the host, are the result of the ability of these hormones to incite, to accelerate, and to mask recognition of many infections. These facts must be kept firmly in mind when these agents are to be used. They are powerful and dangerous substances and must be so treated. There are many gaps in our knowledge of the subject under discussion. Further work is needed to clarify the entire picture.

REFERENCES

1. Abernathy, R The effect of cortisone on experimental brucellosis. J. Clin. Inv., 30: 626, 1951
2. Ainslee, J D, Francis, T, Jr and Brown, G. C ACTH in experimental poliomyelitis in monkeys and mice J Lab & Clin Med., 38: 344-358, 1951.
3. Barrow, J G and Sikes, C R. Decubitus ulcer in rheumatic fever treated with cortisone J A M A, 147: 41, 1951
4. Benditt, E. P and Schiller, S Influence of ACTH upon alteration in capillary permeability induced by hyaluronidase in rats Proc. Soc Exp Biol & Med, 75: 782, 1950
5. Bloch, R. G, Vennesland, M D and Gurney, C. The effect of cortisone on tuberculosis in the guinea pig J. Lab & Clin Med, 38: 133-148, 1951
6. Boling, L, Neukirk, J, Baxter, P, Fargan, S and Kinsell, L W The use of ACTH in the management of generalized peritonitis A preliminary report J Clin End, in press
7. Clark, W. S, Manning, E L and Appleton, J E Observations on the metabolic and anti-inflammatory effects of desoxycorticosterone, testosterone, and cortisone in a patient with rheumatoid arthritis J Clin Inv, 30: 633, 1951
8. [illegible] and Cook, C D [illegible] Conf, J R Mote, [illegible]
9. [illegible] f ACTH and corti- [illegible] -706, 1951
10. [illegible] ichael, M, Jr and Bloom, W L [illegible] v induced peritoneal exudates Bull [illegible]
11. [illegible] influence of cortisone on experimental tuberculosis [illegible], 1951
12. Dougherty, T F. and Schneebeli, G L Role of cortisone in regulation of [illegible] Proc Soc Exp Biol & Med 75: 854, 1950
[illegible] rs de traitment prolongé par (ACTH). Presse Méd, 59: 442, 1951

55 PERLA, D AND MARMORSTON, J.: Natural Resistance and Clinical Medicine. Boston, Little, Brown, and Co , 1941.

56. POPP, C G , OTTOSEN, P. AND BRASHER, C A Cortisone and pulmonary tuberculosis J. A. M A., 147: 241-242, 1951

57. POWELL, A. M , SNYDER, M. J., MINOR, J. V., JR AND BENSON, J F.: The use of terramycin in Rocky Mountain spotted fever Bull Johns Hopkins Hosp , 89: 30, 1951.

58 REDMOND, W. B Influence of cortisone on the natural course of malaria in the pigeon Proc Soc Exp Biol. & Med , 79: 258-261, 1952

59 REEDER, W. H. AND MACKEY, G. S : Nebulized cortisone in bacterial pneumonia. Dis. Chest, 18: 528, 1950.

60 REINMUTH, O M AND SMITH, D T.: The effect of ACTH on pneumonia induced with tuberculin in the lungs of sensitized rabbits Tr. Nat Tub Assn , 34-40, 1951.

61. ROCHE, M : Clinical effects of ACTH in typhoid fever Proc. Second Clin ACTH Conf , 2: 373 Blakiston, 1951.

62 ROSEN, E : Cortisone treatment of trichinosis Am J. Med Sci , 223: 16, 1952

63 SCHMIDTH, L. H AND SQUIRES, W. L : The influence of cortisone on primate malaria J. Exp Med , 94: 501-520, 1951.

64 SCHWARTZMAN, G Enhancing effect of cortisone upon poliomyelitis infection (NEF1) in hamsters and mice Proc Soc Exp Biol & Med , 75: 835, 1950

65 SENSENBACH, C W.: Personal communication

66 SHELDON, W. H., CUMMINGS, M M AND EVANS, L D. Failure of ACTH or cortisone to suppress tuberculin skin reaction in tuberculous guinea pigs Proc Soc Exp Biol & Med , 75: 616-618, 1950

67. SMADEL, J. E , LEY, H L , JR AND DIERCKS, F H : Treatment of typhoid fever I Combined therapy with cortisone and chloramphenicol. Ann Int. Med , 34: 1-10, 1951.

68 SOFFER, L J , SCHWARTZMAN, G , SCHNEIERSON, S S AND GABRILOVE, J L Inhibition of the Schwartzman phenomenon by adrenocorticotrophic hormone (ACTH) from the adenohypophysis Science, 111: 303-304, 1950

69 SPAIN, D M AND MOLOMUT, N : Effects of cortisone on the development of tuberculous lesions in guinea pigs and on their modification by streptomycin therapy Am Rev Tub , 62: 337-344, 1950

70 SPAIN, D. M , MOLOMUT, M AND HABER, A Biological studies on cortisone in mice Science, 112 335-337, 1950

71 SPAIN, D M , MOLOMUT, N AND HABER, A Studies on the cortisone effects on the inflammatory response J Lab & Clin Med , 39: 383-389, 1952

72 STOLTE, J B AND SAS, G J Chloramphenicol and ACTH in smallpox. Lancet, 2: 715, 1951

73 TAUBENHAUS, M AND AMROMIN, G D The effects of the hypophysis, thyroid, sex steroids, and the adrenal cortex upon granulation tissue J Lab & Clin Med , 36: 7-18, 1950

74 THOMAS, L AND GOOD, R A . Bilateral cortical necrosis of kidneys in cortisone treated rabbits following injection of bacterial toxins Proc Soc Exp Biol & Med , 76: 604, 1951

75 THYGESEN, P , GELLER, H O AND SCHWARTZ, A Effect of cortisone on experimental herpes-simplex keratitis of the rabbit Am J Opth , 34: 885, 1951

76 TURNER, T B AND HOLLANDER, D H Cortisone in experimental syphilis. Bull. Johns Hopkins Hosp., 87: 505, 1950

33. KASS, E. H., INGBAR, S. H., LUNDGREN, M. M AND FINLAND, M.: The effect of ACTH and cortisone on pneumococcal and viral infections in the white mouse. J. Lab. & Clin. Med, 37: 780, 1951
34. KIERLAND, R R., O'LEARY, P A, BRUNSTING, L A AND DIDCOCT, J. W.: Cortisone and corticotropin in dermatology J A. M. A, 148: 23, 1952
35. KILBOURNE, E D AND HORSFALL, F. L., JR.. Increased virus in eggs injected with cortisone Proc. Soc. Exp Biol. & Med., 76: 116, 1951.
36. KILBOURNE, E D AND HORSFALL, F L, JR. Lethal infection with Coxsackie virus of adult mice given cortisone Proc Soc Exp Biol. & Med, 77: 135, 1951
37 KING, ERNEST, Q, JOHNSON, J B, BATTEN, G S AND HENRY, W. L.: Tuberculosis following cortisone therapy J A M A., 147: 238–241, 1951
38 KINSELL, L. W Personal communication to author.
39. KLEINSCHMIDT, R. F AND JOHNSTON, J. M. Miliary tuberculosis in a cortisone treated patient case report with autopsy Ann Int. Med., 35: 694–703, 1951.
40 KLIGMAN, A. M, BALDRIDGE, G. D, REBELL, G. AND PILLSBURY, D M: The effect of cortisone on the pathologic response of guinea pigs infected cutaneously with fungi, viruses, and bacteria J Lab & Clin Med, 37: 615–620, 1951
41. KUZELL, W C AND MANKLE, E. A *Cortisone acetate and terramycin* in polyarthritis of rats Proc. Soc Biol & Med, 74: 677, 1950
42 LEMAISTRE, C AND TOMPSETT, R The evolution of tuberculous lesions in the guinea pig during administration of adrenocorticotropic hormone (ACTH) or cortisone Am Rev Tub, 64: 295–307, 1951.
43 LOOSLI, C G, HULL, R B, BERLIN, B S AND ALEXANDER, E R The influence of ACTH on the course of experimental influenza type A virus infection. J Lab & Clin Med, 37: 464–476, 1951
44 LURIE, M B, ZAPPASODI, P, DANNENBERG, A M, JR. AND SWARTZ, I B Constitutional factors in resistance to infection the effect of cortisone on the pathogenesis of tuberculosis Science, 113: 234–237, 1951
45 MICHAEL, M, JR Unpublished observation
46 MICHAEL M, JR The effect of cortisone and ACTH on bacterial infections. Sou Med J, 44: 450–453, 1951
47 MICHAEL, M, JR, CUMMINGS, M M AND BLOOM, W L Course of experimental tuberculosis in the albino rat as influenced by cortisone Proc Soc Exp Biol & Med, 75: 613–616, 1950
48 MICHAEL, M, JR AND WHORTON, C M Delay of early inflammatory response by cortisone Proc Soc Exp Biol & Med, 76 754–756, 1951
49 MIRICK, G S The effect of adrenocorticotropic hormone and cortisone on antibody production in human beings J. Clin Inv, 29: 836, 1950
50 MOGABGAB, W J AND THOMAS, L. The effects of cortisone on experimental infection with Group A hemolytic streptococci in rabbits J Lab & Clin Med, 36: 968, 1950
51. MOGABGAB, W J AND THOMAS, L The effects of cortisone on bacterial infection. J Lab & Clin Med, 39: 271–290, 1952
52 MURRAY, R AND BRANHAM, S E Effect of cortisone and ACTH on adrenals in experimental diphtheria, shiga, and meningococcal infection Proc Soc Exp. Biol & Med, 78: 750, 1951
53 NELSON, J AND GOLDSTEIN, N Nature of the Waterhouse-Friederichsen syndrome J A M A., 146: 1193, 1951
54. NEWMAN, L R. Waterhouse-Friederichsen syndrome Report of a case J A. M A., 146: 1229, 1951.

Prevention of Rheumatic Fever*

CHARLES H. RAMMELKAMP, JR., M.D. AND FLOYD W. DENNY, M.D.

Many attempts have been made to reduce the incidence of acute rheumatic fever. It is not necessary to justify the interest in this problem since it is well known that rheumatic fever is a major cause of death in the young child and rheumatic valvular heart disease is an important cause of chronic disability. Most methods employed for the prevention of rheumatic fever prior to 1939 were of questionable value, but more recently techniques have been developed which may contribute significantly toward the control of this disease. So far effective techniques are based on control of the respiratory infection that precedes rheumatic fever.

The majority of patients with rheumatic fever give a history of a preceding respiratory infection. Haig-Brown (31) in 1886 observed 345 young healthy boys with tonsillitis and 29 subsequently developed heart disease. Subsequent studies have shown that acute rheumatic fever is especially likely to develop following tonsillitis or pharyngitis Bacteriological and immunological investigations have indicated that the precipitating infection is usually, if not invariably, caused by Group A streptococci (13, 64).

Since the natural history of the disease includes contact with group A streptococci, parasitism by the organism (streptococcal infection), development of rheumatic fever, and finally, the subsequent appearance of rheumatic heart disease, it is important for the investigator to evaluate the effect of any procedure tested on each phase of the natural history of the disease For example, surveys to determine the relationship of rheumatic valvular heart disease to different environments may only reflect past experience with group A streptococci of the population studied. Since it is well established that these organisms may attack any population, it is difficult to conclude that any one environmental factor is responsible for an observed effect in a single survey Furthermore, the environmental factor being studied may not have been present at the time of the initial insult (streptococcal infection) which precedes rheumatic valvular heart disease by several years.

The problem of evaluation is exceedingly difficult because few data are available which include information on all aspects of the natural history

* This investigation was conducted under the sponsorship of the Commission on Acute Respiratory Diseases and the Commission on Streptococcal Diseases, Armed Forces Epidemiological Board, and supported by the Office of The Surgeon General, Department of the Army, Washington, D. C

77. VOLLMER, E P.: The course of pneumococcal infections in mice during treatment with antibacterial substances and adrenal cortical extract. J. Inf. Dis, 88: 27, 1951.
78 VOLLMER, E. P. AND HURLBUT, H. S.. Ineffectiveness of cortisone therapy in mice infected with Japanese B encephalitis and the adverse effects of high doses. J. Inf Dis, 89: 103, 1951.
79. VON BRAND, T, TOBIE, E. J. AND MEHLMAN, B.: The failure of cortisone and ACTH to influence gluconeogenesis in trypanosomiasis in the fasting rat. Am J. Hyg., 54: 76, 1951.
80 WHORTON, C M, MICHAEL, M., JR, CUMMINGS, M. M AND BLOOM, W. L.: Unpublished observations
81 WOLF, A, KABAT, E A, BEZER, A. E. AND FOUSECA, J. R. C.. Activation of trypanosomiasis in rhesus monkeys by cortisone Fed Proc., 10: 375, 1951.
82 WOODS, A. C. AND WOOD, R M The action of ACTH and cortisone on experimental ocular inflammations. Bull Johns Hopkins Hosp, 87: 482-504, 1950.
83. WOODWARD, T E., HALL, H E., DIAS-RIVERA, R, HIGHTOWER, J. A., MARTINEZ, E. AND PARKER, R T.. Treatment of typhoid fever. II. Control of clinical manifestations with cortisone Ann. Int. Med, 34: 10-20, 1951.
84. WORKMAN, J B., HIGHTOWER, J A, BORGES, F, FROMAN, E AND PARKER, R. T Cortisone as an adjunct to chloramphenicol in the treatment of Rocky Mountain spotted fever. Presented at Sou Soc. Clin. Res, Atlanta, Ga, Jan 19, 1952.

streptococcal infections and acute rheumatic fever are prone to reach a peak of maximal incidence during the winter and early spring months The effect of geographical location (and therefore altitude, humidity, etc.) appears to be related to the acquisition of the streptococcal infection itself, since once such an infection is established the attack rate of rheumatic fever is relatively constant (53). There is little evidence of racial immunity to rheumatic fever or to rheumatic heart disease. Thus, the fact that inhabitants of Puerto Rico have a low incidence of rheumatic fever on that island but are highly susceptible when transported to New York indicates that the difference is probably due to an increased opportunity to acquire streptococcal infections in New York (13). The observations on rheumatic heart disease by Nichol (49) in the school children in Florida are best explained by a similar mechanism The native-born children had a much lower incidence of rheumatic valvular heart disease than children born in the north and subsequently transported to Florida.

Epidemiological studies show that streptococcal infections, rheumatic fever, and rheumatic heart disease tend to occur in populations living in crowded conditions and poor economic circumstances. Breese (8) has demonstrated in a study of Navy recruits that the amount of respiratory disease is related to the number of individuals housed in a single unit. Likewise, it has been shown that the attack rate for various respiratory infections increases as the size of the family unit increases (21, 42) indicating that the primary factor for the increased rate may be multiple effective exposures within the living unit.

Attempts have been made to eliminate streptococci from the environment by the use of ultraviolet light, aerosols, and oiling floors and blankets but most of these procedures result in little reduction in the incidence of streptococcal infections (55), although occasionally infections appear to be reduced (82). There is no information concerning the effect of such techniques on the incidence of rheumatic fever.

Tonsillectomy

The frequent association of tonsillitis with acute rheumatic fever led to the belief that the tonsils served as a focus of infection in rheumatic fever Although *complete* removal of these organs will obviously prevent tonsillitis, there are no good bacteriological studies which indicate that the incidence of streptococcal respiratory infections is decreased by such operations. Wallace and Smith (66) found that the removal of the tonsils before the age of five years did not protect the child against scarlet fever or rheumatic fever. In fact, they stated that "tonsillectomy performed during early childhood may even increase a child's susceptibility to acute rheumatism."

of the disease. Furthermore, the clinical diagnosis of streptococcal parasitism (infection), rheumatic fever, and rheumatic heart disease is difficult even under ideal circumstances. There are no specific tests for the diagnosis of rheumatic fever. The problem is made difficult also because the streptococcal infection, acute rheumatic fever and even valvular heart disease may be so mild that no or few symptoms are produced Thus the criteria for diagnosis of the several phases of streptococcal infections may vary considerably, so that it is difficult to compare the results of various investigators.

In general, three population groups have been employed to test the efficacy of a given procedure in lowering the incidence of rheumatic fever or rheumatic heart disease. These include the general population, persons who have experienced one or more attacks of rheumatic fever, and patients with the active disease. In the present review the data on prevention of rheumatic fever are re-evaluated considering the effect of each procedure on acquisition of group A streptococci, on the streptococcal infection, on the development of rheumatic fever and on the subsequent appearance of rheumatic heart disease.

PREVENTION OF INITIAL ATTACKS OF RHEUMATIC FEVER

Little effort has been made to prevent initial attacks of rheumatic fever. This is not surprising since the attack rate for rheumatic fever is so low that exceedingly large population groups would have to be included in any study of the prevention of rheumatic fever or rheumatic heart disease. Accurate figures on the incidence of rheumatic fever are not available, but in 1927 Atwater (5) estimated that 0 15 per cent of the population in the United States develops rheumatic fever each year After proved group A streptococcal infections the attack rate is approximately 3 per cent (52) Thus it may be estimated that in a population of 155,000,000 each year there are approximately 7,750,000 streptococcal infections which are followed by 233,000 attacks of rheumatic fever

Environmental Factors

Although there have been numerous epidemiological studies of streptococcal infections, rheumatic fever, and rheumatic heart disease in relation to the effect of various environmental factors, there have been few attempts to alter a single factor under controlled conditions in the general population to determine its effect on the incidence of these diseases. In this review it is impossible to discuss all contributions to the subject, instead, certain factors which apparently alter the attack rates will be presented briefly

Streptococcal respiratory infections, rheumatic fever, and rheumatic heart disease are most prevalent in the temperate zones (74), where both

two or more days to groups of 5,000 men and followed the hospital admissions for respiratory disease. Almost immediately after institution of prophylaxis the incidence of streptococcal infections was greatly reduced and did not return to control levels for over a week after cessation of sulfadiazine administration. These studies also demonstrated that the sulfonamide drugs must be given continuously if such infections are to be prevented.

The Navy (19) experience included observations made on over 600,000 trainees who received 0 5 to 1 gram of sulfadiazine daily. Such prophylaxis resulted in an 85 per cent reduction in the incidence of streptococcal infections and rheumatic fever. The use of 0 5 gram daily appeared to be almost as effective as 1 0 gram and was followed by fewer toxic reactions In general, toxic reactions occurred in approximately 0 01 per cent; and these included granulocytopenia and exfoliative dermatitis. Most of the fatalities resulted from the therapeutic use of sulfadiazine in persons with granulocytopenia due to the prophylactio measures.

The development and spread of sulfonamide-resistant strains was responsible for the abandonment of sulfadiazine prophylaxis in the Armed Services. It should be emphasized, however, that at the time of the report on the first six months' experience, resistant strains had not become a serious problem From these results it would appear entirely reasonable that sulfadiazine might be employed on a more limited scale for the prevention of streptococcal infections and rheumatic fever. If resistance develops penicillin can be substituted as the prophylactic agent.

The sulfonamide drugs have been used extensively in the treatment of streptococcal infections once they become manifest clinically; however, such therapy has little or no effect on the incidence of subsequent attacks of rheumatic fever (20).

Penicillin

Experience with penicillin as a prophylactic agent against rheumatic fever *by prevention of the initial streptococcal infection is limited* In a recent study (48) it was found that the oral administration of 100,000 units of penicillin once daily to naval personnel resulted in a decreased incidence of streptococcal infections. Unpublished studies from the Streptococcal Disease Laboratory have shown that the oral administration of 1,000,000 units twice each day for 10 days eradicates streptococci from carriers and prevents streptococcal infections.

Another approach to the problem of prevention of initial attacks of rheumatic fever has been the treatment of the preceding streptococcal infection. In 1947, Goerner, Massell, and Jones (28) suggested that treatment of such infections with penicillin might prevent recurrent attacks. Kilbourne and Loge (39) observed no cases of rheumatic fever among 29

Selkirk and Mitchell (60) studied 136 children over a three year period after removal of the tonsils and found that colds, nasal obstruction and sore throat were reduced in incidence. Campbell and Warner (12) in a study of 967 children, of whom 124 had had a tonsillectomy, concluded that removal of the tonsils did not reduce the incidence of rheumatic fever or carditis. The extensive study of school children in England reported in 1938 (62) showed that there was no difference between the tonsillectomized and nontonsillectomized children as to the incidence of nasopharyngeal infections, scarlet fever, or rheumatic fever. Although Kaiser (38) reported that the removal of the tonsils resulted in a slight decrease in the incidence of scarlet fever, chorea, rheumatic fever, muscle pains and carditis, it is difficult to interpret the data because the methods are not detailed. In the Armed Services one study (63) has shown that the incidence of tonsillectomized patients with streptococcal infections or rheumatic fever was the same as that in the population from which these groups came.

From these studies there is little evidence that tonsillectomy prevents either streptococcal infections or initial attacks of acute rheumatic fever

Nutrition

That nutrition may play a role in the susceptibility of an individual to the initial attack of acute rheumatic fever has been suggested by several observers Coburn and Moore (14) reported that in New York City children of wealthy parents contracted streptococcal infections without developing rheumatic fever whereas the children of poor parents living only a short distance away developed rheumatic fever following infections with the same serological types of streptococci The authors pointed out that one difference between these two groups was their diets Jackson and his coworkers (36) stated that the majority of children who developed rheumatic fever in their studies were receiving inadequate diets at the time they developed the attack that brought them under observation They found the degree of deficiency of the diet to be correlated with the incidence and degree of heart damage. These studies, however, fail to establish a definitive role of diet in either susceptibility to streptococcal infections or to rheumatic fever

Sulfonamide Prophylaxis

Soon after the demonstration that sulfanilamide exerted an antibacterial effect against β-hemolytic streptococci both *in vitro* and *in vivo,* the drug was employed as a prophylactic agent against streptococcal infections (15, 65). During World War II sulfadiazine was used in an attempt to control streptococcal infections and rheumatic fever among the military population. Hodges (32) administered 1 to 2 grams of sulfadiazine daily for

rheumatic carditis, which is the primary purpose of therapy. Preliminary studies (29) indicate that all electrocardiographic abnormalities are not prevented by the treatment of the streptococcal infection with penicillin. At the present time, however, it should be assumed that adequate therapy probably will prevent carditis. An important aspect of penicillin therapy is the fact that the carrier state is eliminated in the majority of patients. Since the patient convalescent from a streptococcal infection is an important source of new infections, the eradication of streptococci should prevent secondary cases of infection and rheumatic fever.

Aureomycin

Aureomycin has been used in the prevention of rheumatic fever by treating the streptococcal illness. In a small group of 80 patients (10) no instance of rheumatic fever was observed, although there were 5 attacks of rheumatic fever among 198 patients who served as a control group. A subsequent controlled study of 2,039 patients with streptococcal exudative tonsillitis or pharyngitis showed that treatment of the respiratory infection with aureomycin resulted in a 75 per cent reduction in the incidence of subsequent rheumatic fever (34) If aureomycin is employed, therapy should be continued for at least 6 days. This drug does not appear to be as effective as penicillin in eradicating streptococci from the throat.

Cortisone

Because of the reported beneficial effect of cortisone on the clinical course of acute rheumatic fever, this drug was employed by Hahn and his associates (30) in the treatment of streptococcal infections in order to determine its effect on post-streptococcal complications Eighty-seven patients received 5 days of cortisone therapy and 87 patients were given saline and served as controls There were 7 cases of rheumatic fever, 2 of which had received cortisone. Electrocardiographic abnormalities were observed in 8 of the treated group and 7 of the controls.

Immunization

Several attempts have been made to immunize military populations against streptococcal infections Results of the first study (24) indicated that the administration of heat-killed organisms resulted in a slight reduction in the incidence of streptococcus infections A second study employing heat-killed and ultraviolet-killed type 19 and 17 streptococci failed to show any beneficial effect.

Coburn and Pauli (17) observed that recurrent rheumatic attacks were especially likely to occur following infection with strains of streptococci which produced erythrogenic toxin and streptolysin O, and infections

patients treated with 20,000 to 50,000 units of penicillin every three hours for 4 to 7 days; 3 cases among 47 patients receiving a single injection of 300,000 units of aqueous penicillin daily for 7 days; and 2 instances of rheumatic fever among 51 controls. Intensive therapy, as employed in the 29 patients, resulted in a marked inhibition of antibody formation. Since a characteristic feature of the patient who develops rheumatic fever is the production of large amounts of streptococcal antibodies, the results of Kilbourne and Loge indicated that intensive therapy might reduce the incidence of rheumatic fever.

An extensive study of the effect of penicillin therapy of streptococcal infections on the attack rate of rheumatic fever was undertaken among soldiers. In these studies procaine penicillin G in oil containing 2 per cent aluminum monostearate was employed in three different dosage schedules (68) A total of 1,178 patients received penicillin and 1,162 received no penicillin and served as controls. In these two groups 978 patients who received penicillin and 996 controls were followed for post-streptococcal complications. A total of 23 cases of rheumatic fever were observed in the control group and 1 attack occurred in the treated patients within 34 days of the onset of the streptococcal sore throat. These data establish the effectiveness of penicillin therapy of streptococcal infections in the prevention of the clinical manifestations of rheumatic fever. If the patients with rheumatic fever who gave a history indicating previous experience with the disease are excluded, there were 16 instances of initial attacks occurring within 34 days of the onset of sore throat in the control groups and no rheumatic fever in those who received penicillin

These studies also showed that the degree of inhibition of antibody and the degree of effectiveness of therapy in eradicating the streptococcus from the throat were related to the duration of antibacterial therapy Furthermore, there was evidence that the larger doses of penicillin were more effective than smaller doses in decreasing the incidence of post-streptococcal arthralgia and fever As a result it was recommended that patients with streptococcal tonsillitis and pharyngitis should receive at least two injections of 600,000 units of procaine penicillin G in oil containing 2 per cent aluminum monostearate 72 hours apart. Patients with a past history of rheumatic fever or who have evidence of rheumatic valvular heart disease should receive more intensive therapy

These results are at variance with those published by Weinstein, Backrach and Boyer (73) who used penicillin in the treatment of 167 patients with scarlet fever and found that 12 developed acute rheumatic fever. Since a control group of patients was not observed it cannot be concluded that treatment did not alter the attack rate of rheumatic fever These authors pointed out, however, that treatment with penicillin may not prevent

concerning the incidence of recurrent attacks of rheumatic fever. There is evidence that the risk of recurrence is, in part, a function of the age of the individual. Wilson (75) reports that between the ages of 4 and 13, 25 per cent develop recurrences, between 14 and 16, 9 per cent, and between 17 and 25, 4 per cent. Although it has been assumed that age alters susceptibility to rheumatic fever, review of the literature fails to confirm this common belief (53). It would appear that the primary cause for the decreasing recurrence rate as age increases is that the population has fewer effective exposures to group A streptococci. The fact that initial attacks of rheumatic fever appear to be a function of streptococcal infections and independent of age is evidence in support of the above hypothesis (53).

Another factor of importance in relation to the recurrent attack rate is the number of years of freedom from rheumatic fever Wilson and Lubshez have stated that the risk of recurrence in the first year following an attack is twice the risk during the second year, and three times the risk during the third year. Again, this decreasing attack rate may be due to failure to have an effective contact with the streptococcus There is evidence (3, 22, 81) that individuals with a high antibody titer to various streptococcal products experience a higher attack rate of rheumatic fever following a streptococcal infection than individuals with low antibody titers Since the attack of rheumatic fever is in itself evidence of recent contact with group A streptococci, it is not surprising that the recurrence rate decreases as the interval of freedom from rheumatic fever increases.

Environmental Factors

It has been common practice to place individuals with acute rheumatic fever or rheumatic heart disease in general or specialized hospitals Since at the time of onset of rheumatic symptoms at least 60 per cent of patients still harbor group A streptococci in the throat (68), the environment is likely to be contaminated with multiple types of group A streptococci Certainly epidemics of streptococcal infections and rheumatic fever were not uncommon in rheumatic hospitals (61). It is probable that many recurrent attacks might have been avoided in the past by recognition of the increased hazard of rheumatic patients exposed to such contaminated environments.

Prior to the introduction of sulfonamide and penicillin prophylaxis several attempts were made to alter the environment of the rheumatic subject. The fact that rheumatic patients were especially likely to be living under crowded conditions (58) led to the attempt to place such individuals in a new uncrowded environment. Hubbard and Griffin (35) observed no recurrent attacks in a study of 48 rheumatic children placed in an open-air sanatorium Furthermore, there were no streptococci isolated from the

not followed by rheumatic fever frequently were caused by streptococci which failed to produce these toxins. In 1932–33 approximately 100 nurses were immunized with scarlatinal toxin and a similar number served as controls (18). The incidence of streptococcal infections was not significantly different in the two groups, and subsequently there were 2 instances of rheumatic fever and 1 case of nephritis in the treated group, and 1 case of rheumatic fever and 1 case of nephritis in the control group.

It should be emphasized that immunity to streptococcal infection is type specific and such antibodies may not develop for 8 to 10 weeks following infection No information is available as to whether the procedures employed in the above studies were adequate to stimulate type specific immune bodies.

From the recent studies of Wannamaker and Denny (67) it would appear that the presence of bactericidal antibodies does protect against infection with homologous types, but not against heterologous types of streptococci It is possible that immunization with M antigen may produce solid immunity and thereby prevent rheumatic fever. Vaccines thus far employed, however, have been very toxic (54).

PREVENTION OF RECURRENT ATTACKS

In contrast to the statistical problem of evaluation of control procedures in the general population, relatively small groups of individuals who have had one or more attacks of rheumatic fever may be used for study units since the recurrent attack rate is high Thus, in a summary of the reported cases (52) it was found that in a group of 311 patients who had had rheumatic fever, 41 per cent developed a recurrence following an infection with group A streptococci. This is in contrast to a 3 per cent attack rate in the general population experiencing such infections.

Ideally, in order to determine the mechanism whereby experimental techniques alter the recurrent attack rate of rheumatic fever, each subject should be followed by clinical, laboratory (including cultures of the throat), and immunological procedures Such observations would then enable the investigator to determine if the experimental and control patients had been equally exposed to the precipitating agent (streptococci), and if the type of clinical and immunological response was altered after exposure The typing of the infecting streptococcus is likewise important, since occasionally parasitism by some streptococci may not precipitate a recurrent attack (41) The failure to collect information concerning infection with group A streptococci results in difficulties in interpreting data, since experimental and control groups may experience variable numbers of streptococcal infections.

A number of major factors must be considered in the evaluation of data

prevented in their series of 413 rheumatic children. Ash (4) found that in a study of 522 children with rheumatic infections tonsillectomy did not prevent recurrences. She also found that the presence or absence of tonsils did not influence the incidence of cardiac involvement or the death rate. She was very careful to point out that a high incidence of rheumatic exacerbations followed immediately after tonsillectomy performed early in the course of the disease.

In contrast to the above reports Allan and Baylor (2) felt that tonsillectomy and adenoidectomy reduced the incidence of rheumatic recurrences in their patients and recommended this as a form of treatment of rheumatic fever. Kaiser (38) found that recurrences were the same in tonsillectomized and nontonsillectomized patients but that the group who retained their tonsils had a higher mortality than those without. Wilson, Lingg, and Croxford (77) from their studies and those of others state that tonsillectomy for the prevention of rheumatic heart disease is not based on conclusive data.

Nutrition

Coburn and Moore (14) in a study of 50 rheumatic fever patients found that 25 developed a recurrence. In a comparison of these patients with the 25 other patients who did not develop recurrences they found that the diets of the group with recurrences were grossly deficient in protein, iron and vitamin A. Also calcium and riboflavin were found to be low. The authors surmised that rheumatic fever patients who had recurrences were deficient in much of their diet. Coburn (13) has also reported that patients who have had acute rheumatic fever are less susceptible to recurrences, even following streptococcal respiratory infections, if they are given large amounts of egg yolk. Jackson and his co-workers, in a study of the environmental factors as they affect recurrences of rheumatic fever, found a correlation between the diet and recurrences. In these groups social and economic factors in the home were poor. Warner and Winterton (69) found that rheumatic subjects had diets low in animal protein and dairy products. However, the control nonrheumatic children of the same social status also had diets deficient in these same materials. In all studies thus far conducted, no conclusive evidence has been presented which indicates that nutrition alters susceptibility to rheumatic attacks.

Salicylates

Because of the apparent beneficial effect of salicylates on the course of acute rheumatic fever, the drug has been employed by a number of physicians in an attempt to prevent recurrent attacks. Perry (50) gave salicylates continuously after a rheumatic attack and observed 5 relapses among 41

environment in spite of the fact that the organism could be obtained from the oropharynx of these children. Schlesinger (58, 59) noted that recurrences were seldom encountered under open-air conditions in the country It would appear that the effectiveness of such therapy was due to the decreased numbers of group A organisms in the environment.

Rheumatic fever apparently occurs less frequently in tropical and subtropical areas than in the temperate zones. Because of this association several attempts have been made to prevent recurrent attacks by transporting the rheumatic child to southern areas In general, observations have been limited to small groups and sufficient details are not available to allow critical evaluation of the studies. Coburn (13) put 10 New York residents in an open building in San Juan, Puerto Rico, during the winter months and noted that on return to New York symptoms returned. It is to be noted, however, that on return to this country the patients were either sent home or to the general hospital where contact with group A streptococci was undoubtedly great. Jones, White, Roche, Perdue and Ryan (37) studied 26 children sent to southern Florida, but failed to observe much beneficial effect either on the course of the disease or on the incidence of rheumatic recurrences. Robinson and Currens (56) observed 88 rheumatic children for 114 patient years in Miami. There was 1 definite recurrence and 6 mild recurrent attacks. Severe tonsillitis and scarlet fever were not observed. The incidence of *beta*-hemolytic streptococci obtained on culture of the oropharynx remained about 10 to 20 per cent. It is believed that recurrences occur less commonly in the south, but if such therapy is to be effective the rheumatic subject should avoid crowded areas.

There is no doubt that the environment to which the rheumatic patient is exposed should be rendered free of streptococci or the patient should be protected from acquisition of such organisms In rheumatic hospitals this is best obtained by eradicating all group A organisms from the respiratory tract of each new admission. In general hospitals, at school, or at home it is impossible to keep the environment free of streptococci so that continuous sulfonamide or penicillin prophylaxis of the individual patient is required

Tonsillectomy

The conclusions of various investigations concerning the effect of tonsillectomy on the incidence of recurrences of rheumatic fever are conflicting Finland, Robey and Heimann (27) found in a study of 654 patients with acute migratory polyarthritis that the presence or absence of tonsils did not alter the course of the disease or affect the frequency of recurrence. Sheldon (61) found that tonsillectomy had no effect on the incidence of relapses. Wilson, Lingg, and Croxford (77) found that recurrences were not

prevented in their series of 413 rheumatic children Ash (4) found that in a study of 522 children with rheumatic infections tonsillectomy did not prevent recurrences. She also found that the presence or absence of tonsils did not influence the incidence of cardiac involvement or the death rate. She was very careful to point out that a high incidence of rheumatic exacerbations followed immediately after tonsillectomy performed early in the course of the disease.

In contrast to the above reports Allan and Baylor (2) felt that tonsillectomy and adenoidectomy reduced the incidence of rheumatic recurrences in their patients and recommended this as a form of treatment of rheumatic fever. Kaiser (38) found that recurrences were the same in tonsillectomized and nontonsillectomized patients but that the group who retained their tonsils had a higher mortality than those without Wilson, Lingg, and Croxford (77) from their studies and those of others state that tonsillectomy for the prevention of rheumatic heart disease is not based on conclusive data.

Nutrition

Coburn and Moore (14) in a study of 50 rheumatic fever patients found that 25 developed a recurrence. In a comparison of these patients with the 25 other patients who did not develop recurrences they found that the diets of the group with recurrences were grossly deficient in protein, iron and vitamin A. Also calcium and riboflavin were found to be low The authors surmised that rheumatic fever patients who had recurrences were deficient in much of their diet Coburn (13) has also reported that patients who have had acute rheumatic fever are less susceptible to recurrences, even following streptococcal respiratory infections, if they are given large amounts of egg yolk Jackson and his co-workers, in a study of the environmental factors as they affect recurrences of rheumatic fever, found a correlation between the diet and recurrences In these groups social and economic factors in the home were poor Warner and Winterton (69) found that rheumatic subjects had diets low in animal protein and dairy products However, the control nonrheumatic children of the same social status also had diets deficient in these same materials In all studies thus far conducted, no conclusive evidence has been presented which indicates that nutrition alters susceptibility to rheumatic attacks

Salicylates

Because of the apparent beneficial effect of salicylates on the course of acute rheumatic fever, the drug has been employed by a number of physicians in an attempt to prevent recurrent attacks Perry (50) gave salicylates continuously after a rheumatic attack and observed 5 relapses among 41

children, whereas there were 19 relapses among 106 control patients. It was concluded that salicylates did not prevent relapses. This experience was similar to that reported by Miller (47).

Another approach has been the administration of salicylates at the onset of the streptococcal sore throat. Such treatment is continued for a period of at least 4 weeks. Coburn and Moore (16) observed 1 recurrence among 47 patients receiving such treatment whereas there were 57 recurrent attacks in 186 control patients Schlesinger (59) reports that such therapy prevented serious relapses; however, Sheldon (61) observed 3 recurrences among 7 patients treated with salicylates. It is possible that salicylates used in this manner merely mask some of the symptoms of rheumatic fever but do not affect the occurrence of rheumatic carditis.

TABLE I

*Effect of Sulfonamide Prophylaxis on the Recurrence of Rheumatic Fever**

CONTROL			SULFONAMIDE PROPHYLAXIS		
Patient seasons	Rheumatic attacks		Patient seasons	Rheumatic attacks	
	Number	Per cent		Number	Per cent
1739	241	13 9	1447	27	1 9

* These data include the summary of Dodge (23) and studies of Baldwin (6), Rubbo (57), and Feldt (26)

Sulfonamides

Coburn and Moore (15) and Thomas and France (65) in 1939 reported that the daily administration of sulfanilamide to rheumatic subjects prevented recurrent attacks by preventing the preceding streptococcal respiratory infection. Subsequent to these reports a number of investigations were instituted. The experience is summarized in table 1 In these studies either sulfanilamide or sulfadiazine was administered in doses of 0.5 to 2 0 grams daily. Such therapy resulted in an 85 per cent reduction in the recurrent attack rate of rheumatic fever With the exception of 1 death from agranulocytosis and a few individuals who did not tolerate the drug, therapy was continued in all patients At the present time sulfadiazine is commonly employed as the prophylactic agent in daily dosage of 0.5 to 1 gram daily.

The development and dissemination of resistant strains of group A streptococci have never been serious problems in prophylaxis of individual rheumatic subjects *It is to be emphasized, however, that such therapy does not prevent all streptococcal infections and recurrent attacks of rheumatic fever.* Whether strains of streptococci which cause infection in patients are resistant to the bacteriostatic action of the drug has not been determined.

The question as to what patients should receive prophylaxis and how long such treatment should be maintained has never been answered adequately. It is assumed that repeated attacks are likely to cause increased damage to the heart, so that there is no argument about the desire to prevent recurrences. *It would appear entirely reasonable to insist that every person who has had rheumatic fever with signs of carditis should receive the benefit of prophylaxis for a minimal period of two years.* In most instances such preventive measures should continue for 3 to 5 years. In arriving at a decision as to how long such prophylaxis should continue, one should consider the degree of damage to the heart existing at the start of prophylaxis, the opportunities (risk) of acquiring a streptococcal infection, and the period of freedom from the last attack of rheumatic fever or a streptococcal infection. In this regard, the young child, age 5 to 10 years, who lives in a crowded home and attends a crowded school, requires steady protection.

Penicillin

In 1947 Goerner, Massell and Jones (28) showed that *beta*-hemolytic streptococci could be eradicated from the throat of the majority of carriers by the application of penicillin for 10 days. Since that time there have been several reports concerning the control of streptococcal infections with penicillin in patients who have had acute rheumatic fever. Burke (11) observed no recurrences of rheumatic fever in 10 patients receiving 500 unit troches three times a day. One recurrence occurred in the control group of the same size. Malner and Amsterdam (44) in 1947 and Malner (43) in 1950 reported the use of penicillin troches of 1000 and 5000 units each three times a day. In the first report there were no recurrences in 22 patients while four of 22 control patients suffered recurrences. In the second report none of 30 patients suffered recurrences while two of 33 control patients had recurrences.

Another approach to this problem has been the oral administration of penicillin in doses large enough to inhibit the growth of the streptococcus and eradicate the carrier state. Hofer (33) by using 200,000 units in two divided doses prevented streptococcal infections for seven months in 63 children who had had rheumatic fever. Only one patient in this group was found to be a confirmed carrier of group A streptococcus. In the control group of 64 children there were 4 cases of streptococcal respiratory disease and 11 confirmed carriers, all of group A streptococcus.

Kohn, Milzer and MacLean (40) studied 126 children who had had rheumatic fever and found that they could reduce the carrier rate for hemolytic streptococci and recurrences of rheumatic fever by giving penicillin by mouth. They studied various treatment schedules but did not

administer penicillin continuously at any time. The majority of the patients were treated with approximately 1,000,000 units a day in 4 or 5 divided doses.

Massell (45) reported that there have been no streptococcal respiratory infections or rheumatic fever recurrences at the Good Samaritan Hospital over a five year period with the prophylactic use of 100,000 or 200,000 units of penicillin orally three times daily.

Evans (25) studied 155 patients with rheumatic fever who were treated with 100,000 units of penicillin by mouth daily before breakfast. One hundred forty-five untreated patients were controls. The carrier state for group A streptococci was greatly reduced. There was only one group A streptococcal infection and no relapses of rheumatic fever in the treated group, while in the control group there were 7 streptococcal infections and 4 relapses of rheumatic fever

Since sulfonamide prophylaxis is not completely effective in preventing recurrences, it is possible that eventually penicillin prophylaxis may be employed Further information is required as to dosage and type of penicillin preparation employed. There is no information concerning the prophylactic use of other antibiotics in the prevention of recurrent attacks.

If streptococcal infections are recognized early in the rheumatic subject, adequate treatment with penicillin will prevent many of the recurrences. Thus, Massell, Sturgis, Knobloch, Streeper, Hall, and Norcross (46) observed only two recurrent attacks after 34 streptococcal infections in rheumatic subjects In contrast, there were 6 recurrences after 12 infections in control subjects It is recommended that penicillin therapy of such patients be instituted immediately and be continued for approximately 10 days It is probably desirable to give as much as 600,000 units of procaine penicillin G each day for the 10 day period.

Massell (45) has noted that treatment of streptococcal infections should not be a substitute for the use of continuous prophylactic measures in rheumatic subjects. This statement is justified, for approximately 40 per cent of streptococcal infections produce few, if any symptoms, and thus recurrences will occur in the rheumatic subjects protected only by treatment of the infection itself

Immunization

Since it was recognized that rheumatic fever followed infection with the streptococcus, a number of investigators have attempted to prevent recurrences by immunization procedures In 1930 and 1931 Wilson and Swift (79) reported the effect of the administration of a heat-killed vaccine on the recurrence of acute rheumatic fever In these preliminary reports it was found that 45 per cent of the immunized subjects and 18 per cent of

the controls remained free of recurrences Although these results were encouraging, follow-up studies (76) over a three year period showed that only 19 per cent of the treated and 17 per cent of the control groups failed to experience recurrences. Since the administration of typhoid vaccine also caused a slight reduction in recurrences, it was concluded that immunization with heat-killed streptococci probably did not influence the incidence of rheumatic recurrences

Favorable results of immunization of rheumatic subjects have been reported by Wasson and Brown (70). In a study extending over several years they found that the administration of filtrates of a culture of NY5 strain of hemolytic streptococci, or of a tannic acid precipitated toxin of the same strain, the incidence of recurrences was reduced when compared to a control group These data are difficult to interpret since there is insufficient information concerning the comparability of the experimental and control group Furthermore, since such immunization procedures fail to induce type specific immunity, it seems unlikely that streptococcal infections were prevented. The fact that Coburn and Pauli (18) employed toxin from the same strain and failed to prevent initial attacks of rheumatic fever indicates that the favorable results of Wasson may have been fortuitous. Coburn and Pauli (18) report that the administration of NY5 antiserum to rheumatic subjects immediately following a streptococcal infection failed to prevent recurrences Ten patients were given this serum, and 6 developed rheumatic fever.

PREVENTION OF RHEUMATIC HEART DISEASE BY TREATMENT OF RHEUMATIC FEVER

At the present time there is no conclusive evidence that any form of therapy of the acute attack of rheumatic fever reduces the incidence of subsequent rheumatic valvular heart disease. It is true that certain drugs such as salicylates, ACTH and cortisone alter the clinical course of the disease, but it has not been demonstrated that rheumatic carditis is likewise favorably altered

The problem of evaluating the effectiveness of any method of treatment on either rheumatic carditis or valvular heart disease is especially difficult because of the variable course of the illness. The careful studies of Bland and Jones (7) are helpful in this regard, but unless all subsequent streptococcal infections are prevented after the observed attack of rheumatic fever, it may be impossible to determine the effect of any specific therapy.

CONCLUSIONS

Today the most effective method available for the prevention of initial attacks of acute rheumatic fever is the treatment of the preceding strep-

tococcal infection with either penicillin or aureomycin. It should be emphasized, however, that although such therapy prevents the rheumatic attack it has not been established that such therapy prevents rheumatic carditis. Until studies demonstrate this possibility it is advisable to treat all streptococcal infections. These infections may be recognized in many instances since the disease usually begins suddenly with soreness on swallowing, feverishness, and other constitutional symptoms. The lymph nodes at the angle of the jaw usually are enlarged and tender. The oropharynx shows diffuse redness and swelling of the tissues and in many patients exudate is observed. The total leukocyte count is elevated and streptococci in large numbers can be obtained on culture. Using such criteria, it is estimated that approximately 50 to 60 per cent of all streptococcal infections may be recognized and treatment instituted.

In population groups such as in military establishments sulfonamide drugs may be employed on a limited scale for the prevention of streptococcal infections and rheumatic fever Since resistant strains may appear during such therapy, it is likely that penicillin may replace the sulfonamides as a prophylactic agent in the general population.

After rheumatic fever has developed, the patient should be protected from subsequent infections by group A streptococci. If the patient is put in a rheumatic hospital, treatment with penicillin should be instituted in order to eradicate the carrier state before exposure to other rheumatic subjects. If the patient is kept at home or in a general hospital, sulfadiazine or penicillin should be administered daily to protect the patient from acquiring new group A organisms. Any respiratory infection developing in the rheumatic subject should be investigated immediately If the infection is caused by group A streptococci, adequate penicillin therapy should be instituted

The use of other procedures in the prevention of streptococcal infections is of questionable value. Theoretically, putting the rheumatic subject in a streptococcal-free environment is advisable, but practically can seldom be obtained without great sacrifice. It is possible that in the future specific immunization against streptococcal infections may be effective in preventing streptococcal infections and rheumatic fever under certain circumstances.

BIBLIOGRAPHY

1 Adenoidectomy. Am J Dis Child, 42: 9–41, 1931
2 Allan, W. B. and Baylor, J W The influence of tonsillectomy upon the course of rheumatic fever and rheumatic heart disease Johns Hopkins Hosp Bull, 63: 111–123, 1938
3. Anderson, H. C, Kunkel, H G and McCarty, M Quantitative antistreptokinase studies in patients infected with group A hemolytic streptococci A

comparison with serum antistreptolysin and gamma globulin levels with special reference to the occurrence of rheumatic fever J. Clin Invest, 27: 425-434, 1948

4. Ash, R.: Influence of tonsillectomy on rheumatic infection Am J Dis Child, 55: 63-78, 1938

5 Atwater, R. M · Studies in the epidemiology of acute rheumatic fever and related diseases in the United States, based on mortality statistics Am J Hyg, 7: 343-369, 1927.

6 Baldwin, J. S: Sulfadiazine prophylaxis in children and adolescents with inactive rheumatic fever. J. Pediat, 30: 284-288, 1947

7. Bland, E. F. and Jones, T. D Rheumatic fever and rheumatic heart disease A twenty year report on 1000 patients followed since childhood Circulation, 4: 836-843, 1951.

8. Breese, B B., Stanbury, J, Upham, H, Calhoun, A. J, Van Buren, R L and Kennedy, A S.. Influence of crowding on respiratory illness in a large naval training station. War Med, 7: 143-146, 1945.

10. Brink, W. R, Rammelkamp, C. H, Jr, Denny, F W and Wannamaker, L. W.: Effect of penicillin anc aureomycin on natural course of streptococcal tonsillitis and pharyngitis Am J Med, 10: 300-308, 1951

11. Burke, P J Penicillin prophylaxis in acute rheumatism Lancet, 1: 255-256, 1947

12 Campbell, M and Warner, E C: A study of rheumatic disease in children. Lancet, 1: 61-66, 1930

13. Coburn, A F · The Factor of Infection in the Rheumatic State. Baltimore, The Williams & Wilkins Co, 1931

14 Coburn, A F and Moore, L V Nutrition as a conditioning factor in the rheumatic state Am J Dis. Child, 65: 744-756, 1943

15 Coburn, A F. and Moore, L V The prophylactic use of sulfanilamide in streptococcal respiratory infections, with especial reference to rheumatic fever J Clin Invest, 18: 147-155, 1939.

16 Coburn, A. F and Moore, L V Salicylate prophylaxis in rheumatic fever J Pediat, 21: 180-183, 1942

17 Coburn, A F and Pauli, R H Studies on the immune response of the rheumatic subject and its relationship to activity of the rheumatic process IV. Characteristics of strains of hemolytic streptococcus, effective and noneffective in initiating rheumatic activity J Clin Invest, 14: 755-762, 1935

18 Coburn, A F and Pauli, R H Studies on the immune response of the rheumatic subject and its relationship to activity of the rheumatic process V Active and passive immunization to hemolytic streptococcus in relation to the rheumatic process J Clin Invest, 14: 763-768, 1935

19 Coburn, A F and Young, D C The Epidemiology of Hemolytic Streptococcus Baltimore, The Williams & Wilkins Co, 1949

20 Commission on Acute Respiratory Diseases A study of a food-borne epidemic of tonsillitis and pharyngitis due to β-hemolytic streptococcus, type 5. Bull Johns Hopkins Hosp, 77: 143-210, 1945

21 Commission on Acute Respiratory Diseases Unpublished observations

22 Denny, F W, Wannamaker, L W and Rammelkamp, C H, Jr The relation of antibody production to the development of rheumatic fever Am J Dis Child, 80: 506, 1950

23. Dodge, K G, Baldwin, J S and Weber, M. W · The prophylactic use of

sulfanilamide in children with inactive rheumatic fever. J. Pediat , 24: 483–501, 1944

24. Epidemiology Unit No 22 Failure of type specific streptococcus pyogenes vaccine to prevent respiratory infections Naval Med. Bull , 46: 709–718, 1946.

25. Evans, J. A. P Oral penicillin in the prophylaxis of streptococcal infection and rheumatic relapse. Proc. Roy. Soc Med , 43: 206–208, 1950.

26 Feldt, R. H Sulfanilamide as prophylactic measure in recurrent rheumatic infection. Controlled study involving 130 "patient seasons". Am. J Med Sc., 207: 483–488, 1944.

27. Finland, M , Robey, W H. and Heimann, H.: The effect of tonsillectomy on the occurrence and course of acute polyarthritis. Am. Heart J , 8: 343-356, 1933

28 Goerner, J R , Massell, B F and Jones, T. D.: Use of penicillin in the treatment of carriers of β-hemolytic streptococci among patients with rheumatic fever New Eng J Med , 237: 576–580, 1947.

29. Hahn, E O , Eckhardt, G. C. and Stowens, D : The relationship of post-strepcococcal electrocardiographic abnormalities to rheumatic fever and their *reduction by antibiotic therapy Am. J Med , to be published*

30 Hahn, E O , Houser, H B , Rammelkamp, C. H., Jr , Denny, F. W. and Wannamaker, L W Effect of cortisone on acute streptococcal infections and post-streptococcal complications J. Clin Invest , 30: 274–281, 1951.

31. Haig-Brown, C · Tonsillitis in Adolescents. London, Bailliere, Tindall & Cox, 1886

32 Hodges, R G The use of sulfadiazine as a prophylactic against respiratory disease. New Eng J Med , 231: 817–820, 1944

33 Hofer, J W. Oral penicillin for children with rheumatic fever J Pediat , 35: 135–144, 1949.

34 Houser, H B , Eckhardt, G. C , Hahn, E O , Denny, F W , Wannamaker, L W and Rammelkamp, C. H , Jr To be published

35. *Hubbard, J P and Griffin, W A Open-air sanatorium care for patients with* rheumatic fever and rheumatic heart disease New Eng J Med , 223: 968–972, 1940

36. Jackson, R L , Kelly, H G , Robert, C H and Duane, J M Rheumatic fever recurrences in children without sulfonamide prophylaxis An evaluation *of environmental factors* J Pediat , 31: 390–402, 1947

37 Jones, T D , White, P. D , Roche, C F , Perdue, J J and Ryan, H A The transportation of rheumatic fever patients to a subtropical climate J. A. M A , 109: 1308–1309, 1937

38. Kaiser, A D Tonsils in development of the child J A M A , 116: 1151–1156, 1940

39 Kilbourne, E. D. and Loge, J P. The comparative effects of continuous and [illegible]

[illegible] of rheumatic fever J A M A 144 [illegible], 1950

[illegible] he effect of streptococcal [illegible] A three year study J Clin. Invest , 20: 273–287, 1941

42 Lidwell, O. M. and Sommerville, T Observations on the incidence and distri-

bution of the common cold in a rural community during 1948 and 1949. J. Hyg., 49: 365-381, 1951.
43. Maliner, M M.: Oral penicillin in the prophylaxis of recurrent rheumatic fever. J Pediat., 37: 858-861, 1950.
44 Maliner, M M. and Amsterdam, S D.: Oral penicillin in the prophylaxis of recurrent rheumatic fever. J. Pediat., 31: 658-661, 1947.
45. Massell, B. F.: Present status of penicillin prophylaxis of rheumatic fever. Mod. Conc Cardiovascular dis, 20: 108-109, 1951.
46. Massell, B F., Sturgis, G. P., Knobloch, J. D, Streeper, R. B, Hall, T. N., and Norcross, P.: Prevention of rheumatic fever by prompt penicillin therapy of hemolytic streptococcic respiratory infections. Progress report. J. A. M. A, 146: 1469-1474, 1951.
47 Miller, R.: The "specific" use of salicylate in acute rheumatism A consideration of practical objections. Quart. J. Med., 6: 519-540, 1913.
48 Naval Medical Research Unit No. 4, Research Project NM 005 051 15 01. Unpublished observations
49 Nichol, E. S: Rheumatic heart disease in southern Florida. Am Heart J., 9: 63-71, 1933.
50 Perry, C. B.. The value of salicylates in the prevention of rheumatic relapses. Lancet, 1: 861-862, 1933.
51 Prevention of Respiratory Tract Bacterial Infections by Sulfadiazine Prophylaxis in the United States Navy. Navmed 284, Bureau of Medicine and Surgery, Navy Department, Washington.
52 Rammelkamp, C. H, Jr, Denny, F. W. and Wannamaker, L. W.: Epidemiology of rheumatic fever. To be published.
53. Rammelkamp, C H, Jr, Wannamaker, L W and Denny, F. W.: The epidemiology and prevention of rheumatic fever. Bull New York Acad. Med., 28: 321-334, 1952.
54 Rantz, L. A, Randall, E. and Rantz, H. H.: Immunization of human beings with group A hemolytic streptococci. Am. J. Med, 6: 424-432, 1949.
55 Report of the Subcommittee for the Evaluation of Methods to Control Air-borne Infections, of the Committee on Research and Standards· The present status of the control of air-borne infections A. J. P. H, 37: 13-22, 1947
56 Robinson, J J and Currens, J H: Preliminary observations on some children with rheumatic heart disease transported to a subtropical climate. J. Pediat, 28. 426-428, 1946
57 Rubbo, S. D, Holmes, M C. and Stokes, H L Prophylactic sulfanilamide in rheumatic fever. Review of 548 cases Lancet, 2: 311-316, 1949.
58 Schlesinger, B The public health aspect of heart disease in childhood I Lancet, 1: 593-599, 1938
59 Schlesinger, B: The public health aspect of heart disease in childhood II Lancet, 1: 649-654, 1938.
60 Selkirk, T K and Mitchell, A. G.. Evaluation of the results of tonsillectomy and adenoidectomy. Am. J Dis Child, 42: 9-41, 1931.
61 Sheldon, W On acute rheumatism following tonsillitis Lancet, 1: 1337-1341, 1931.
62 Special Report Series No 227 Medical Research Council: Epidemics in Schools London, His Majesty's Stationery Office, 118-125, 1938
63 Streptococcal Disease Laboratory: Unpublished observations

sulfanilamide in children with inactive rheumatic fever. J. Pediat., **24**: 483–501, 1944

24 Epidemiology Unit No. 22: Failure of type specific streptococcus pyogenes vaccine to prevent respiratory infections. Naval Med. Bull., **46**: 709–718, 1946

25 Evans, J A. P.. Oral penicillin in the prophylaxis of streptococcal infection and rheumatic relapse. Proc Roy. Soc. Med., **43**: 206–208, 1950

26 Feldt, R. H Sulfanilamide as prophylactic measure in recurrent rheumatic infection Controlled study involving 130 "patient seasons". Am J. Med Sc, **207**: 483–488, 1944

27. Finland, M, Robey, W H and Heimann, H: The effect of tonsillectomy on the occurrence and course of acute polyarthritis Am Heart J, **8**: 343–356, 1933.

28 Goerner, J R, Massell, B. F and Jones, T. D: Use of penicillin in the treatment of carriers of β-hemolytic streptococci among patients with rheumatic fever New Eng J Med, **237**: 576–580, 1947

29. Hahn, E O, Eckhardt, G C. and Stowens, D: The relationship of post-strepcococcal electrocardiographic abnormalities to rheumatic fever and their reduction by antibiotic therapy Am J Med, to be published

30 Hahn, E O, Houser, H. B, Rammelkamp, C H, Jr, Denny, F. W. and Wannamaker, L. W Effect of cortisone on acute streptococcal infections and post-streptococcal complications J. Clin Invest, **30**: 274–281, 1951

31. Haig-Brown, C Tonsillitis in Adolescents London, Baillière, Tindall & Cox, 1886

32 Hodges, R G The use of sulfadiazine as a prophylactic against respiratory disease New Eng J Med, **231**: 817–820, 1944.

33 Hofer, J W Oral penicillin for children with rheumatic fever J Pediat, **35**. 135–144, 1949

34 Houser, H B, Eckhardt, G C, Hahn, E O, Denny, F. W, Wannamaker, L W and Rammelkamp, C H, Jr. To be published

35 Hubbard, J. P and Griffin, W A Open-air sanatorium care for patients with rheumatic fever and rheumatic heart disease New Eng J Med, **223**: 968–972, 1940

36 Jackson, R L, Kelly, H G, Rohert, C H and Duane, J M Rheumatic fever recurrences in children without sulfonamide prophylaxis An evaluation of environmental factors J Pediat, **31**: 390–402, 1947.

37 Jones, T D, White, P D, Roche, C F, Perdue, J J and Ryan, H A The transportation of rheumatic fever patients to a subtropical climate J. A. M A, **109**: 1308–1309, 1937

38 Kaiser, A D Tonsils in development of the child J A M A, **115**: 1151–1156, 1940

39 Kilbourne, E D and Loge, J P The comparative effects of continuous and intermittent penicillin therapy on the formation of antistreptolysin in hemolytic streptococcal pharyngitis J Clin Invest, **27**: 418–424, 1948

40 Kohn, K H, Milzer, A and MacLean, H Oral penicillin prophylaxis of rheumatic fever. J. A. M. A., 142: 20–25, 1950

streptococcal
year study J

nce and distri

Amebiasis

The Clinician's Responsibility

HENRY E. HAMILTON, M D.

INTRODUCTION

Infection with *Endamoeba histolytica* is widespread in temperate zones as well as in the tropics. Indeed, it is world-wide in distribution, following man wherever he has settled For many years expert clinical, pathological and therapeutic knowledge of amebiasis has been available; yet in pointing up the problem of this disease D'Antoni (19) made the following statement: "The majority of physicians know little about amebiasis and what knowledge they have is usually incorrect The disease is a national as well as a personal problem and one which is apparently becoming more serious." This widespread ignorance of amebiasis has arisen from two major causes: 1) failure of proper dissemination of the known facts of amebiasis, and 2) existence of unsolved problems which have a direct bearing on the clinical approach to this disorder.

It would be trite to mention the incidence and manifestations of amebiasis to workers in the field, yet the practicing physician still believes that amebic infection is a dysentery-like disease confined primarily to people in tropical countries. As has been pointed out clearly by Craig (15, 16) it is a serious error to consider this disease as a dysentery. It is a disease resulting from the invasion of one or more of many tissues of the body and is capable of producing an untold number of divergent symptoms. Thus an infestation with the parasite *Endamoeba histolytica* is properly referred to as amebiasis and the involvement of the liver, bowel, skin or brain is but an aspect of the disease In spite of this concept of amebiasis a review of articles, autopsy reports and hospital records indicates that all too often the discovery of trophozoites of *Endamoeba histolytica* obtained from a tender right upper quadrant abdominal mass, right sided pleural fluid, or from the autopsied bowel in "carcinoma of the intestine" comes as a complete surprise.

Exact figures are not available for the incidence of amebiasis The admittedly incomplete U S. Public Health reports show that 41,781 people had amebiasis from 1933 to 1947 and that during this period 3,305 persons died from the disorder. This mortality figure is serious when it is known that the treatment for amebiasis usually is successful On the basis of surveys compiled from the literature by Craig and Faust (18) it is conservatively estimated that 10 per cent of the population of the United States harbor the parasite *E. histolytica*. All members of this segment of

64. SWIFT, H F.: The etiology of rheumatic fever. Ann. Int. Med., **31**: 715–738, 1949.
65. THOMAS, C. B. AND FRANCE, R.: A preliminary report of the prophylactic use of sulfanilamide in patients susceptible to rheumatic fever. Bull Johns Hopkins Hosp , **64**: 67–77, 1939
66. WALLACE, H. L. AND SMITH, A. B.: The effect of early tonsillectomy on the incidence of acute rheumatism. Edinburgh M J , **43**: 452–457, 1936
67. WANNAMAKER, L. W. AND DENNY, F. W.. To be published
68 WANNAMAKER, L W., RAMMELKAMP, C. H., JR , DENNY, F. W , BRINK, W. R , HOUSER, H B., HAHN, E. O. AND DINGLE, J H : Prophylaxis of acute rheumatic fever by treatment of the preceding streptococcal infection with various amounts of depot penicillin Am J. Med , **10**: 673–695, 1951.
69. WARNER, E C. AND WINTERTON, F. G · A dietetic study of cases of juvenile rheumatic disease. Quart. J Med., **4**: 227–246, 1935
70 WASSON, V P AND BROWN, E E · Further studies in immunization against rheumatic fever Am Heart J , **23**: 291–305, 1942.
71. WASSON, V P AND BROWN, E. E : Immunization against rheumatic fever J. Pediat , **23**: 24–30, 1943.
72. WASSON, V P AND BROWN, E. E : Immunization against rheumatic fever with hemolytic streptococcus filtrate. Am. Heart J., **20**: 1–11, 1940
73 WEINSTEIN, L , BACKRACH, L AND BOYER, N H *Observations on the development of rheumatic fever and glomerulonephritis in cases of scarlet fever treated with penicillin* New Eng J Med , **242**: 1002–1010, 1950
74 WILSON, M G Rheumatic Fever Studies of the Epidemiology, Manifestations, Diagnosis, and Treatment of the Disease During the First Three Decades New York, Commonwealth Fund, 1940.
75. WILSON, M G Susceptibility of the host in rheumatic fever M Clin North Am , **30**: 534–539, 1946
76 WILSON, M G , JOSEPHI, M G AND LANG, D M Intravenous vaccination with streptococci Its influence on the incidence of recurrence of rheumatic fever in children Am J Dis Child , **46**: 1329–1337, 1933
77. WILSON, M G , LINGG, C AND CROXFORD, G Statistical studies bearing on problems in the classification of heart disease IV Tonsillectomy in its relation to the prevention of rheumatic heart disease Am Heart J , **4**: 197–209, 1928
78 WILSON, M G AND LUBSHEZ, R Recurrence rates in rheumatic fever, the evaluation of etiologic concepts and consequent preventive therapy J A M A , **126**: 477–480, 1944
79 WILSON, M G AND SWIFT, H F Influence of intravenous vaccination with streptococci on the prevention of relapses in children with rheumatic fever Tr Assoc Am Phys , **45**: 260–263, 1930
80 WILSON, M G AND SWIFT, H F Intravenous vaccination with hemolytic [illegible] fever in
81 [illegible] sis of [illegible] natic
82 [illegible] epto-

when amebiasis is searched for. The discovery is made then, not by a chance laboratory test, but because the possibility is recognized that certain symptoms and signs could be caused by an amebic infection.

Thus since there is a high incidence of unrecognized amebiasis to which mass survey methods are not readily applicable and because the treatment of this disease is highly satisfactory, the amebiasis problem for the practicing physician is the clinical recognition of the disorder. At the present time, therefore, a positive attack on the amebiasis problem must be made in the doctor's office and at the bedside. It is the objective of this paper to consider the fundamental facts of amebiasis as they relate to the recognition and treatment of this disease as seen in the individual patient Public health and preventative measures will not be discussed

PATHOLOGY

Understanding the variable clinical course of amebiasis has its basis in the interpretation of the pathological findings which have been reviewed critically (16, 18). An infection with *E. histolytica* is possible only after ingestion of the cystic form of the parasite The active trophozoitd or vegetative stage is destroyed by the gastric juice Food or drink has usually been contaminated by flies, food handlers or by direct contact from human feces, through a communication with a sewage system or by the use of night soil for fertilization of crops. Dust is not a source of infection, for cysts of *E. histolytica* die upon drying Once the cyst reaches the terminal ileum and large bowel excystation takes place and the eight motile trophozoites of *E histolytica* are formed The trophozoite probably is unable to live in the lumen of the bowel and must gain access to the tissues of the host to survive Superficial erosions of the mucosa are produced by a cytolytic activity attributed specifically to the parasite. It then invades the tissues locally and establishes colonies of amebae Faust (28) demonstrated these lesions in "healthy carriers" who died accidentally The primary lesion of intestinal amebiasis consists of cytolysis and necrosis of the cells with colonization of the parasites within the involved tissues There is a conspicuous absence of bacterial invasion or an inflammatory reaction about the immediate zone of invasion

The extension of the process may be limited by the repair mechanism of the body and at times spontaneous recovery probably takes place. On the other hand, the process of cytolysis and necrosis may extend, producing deep-seated ulcers of the bowel. In large advanced lesions secondary bacterial invasion occasionally may take place contributing to further pathological changes and clinical symptoms. Extensive granulomatous lesions may be produced especially in the cecal region. With extension of the infection, the deep lymphatics, capillaries, and venules may be in-

the population are considered to have amebiasis because it is thought that cysts in the stool come from bowel ulcers caused by motile trophozoites The clinician rightfully may ask how many persons have symptoms or will have symptoms. Are the organisms found really pathogenic? On the basis of autopsy material and clinical data it is thought that some persons recover from the mild infection spontaneously; thus it is not known to what extent symptoms occur in those who pass cysts On the other hand, careful questioning of many cyst passers reveals they periodically have symptoms compatible with clinical amebiasis. Furthermore, we know that cysts from asymptomatic cyst passers may produce an acute amebiasis when fed to human volunteers. From the available data it is known that some cyst passers may develop serious manifestations of amebiasis and that cysts of *E. histolytica* are potentially dangerous when ingested by a susceptible host.

The laboratory diagnosis of amebiasis has been an almost unsurmountable task for the practicing physician First of all, the methods for obtaining and examining specimens have been time-consuming In many of the clinical forms of the disease encountered in this country very few or no parasites are found in the stool. Frequently the parasites are atypical and difficult to classify. On the other hand, in the tropics and in Korea where our soldiers have become ill with amebiasis the stool specimens have been literally filled with the classical forms of the parasite (34) To add to the difficulties of diagnosis, in this country we lack trained technicians who can recognize the parasite. Even the expert parasitologists sometimes have difficulty in identifying *E histolytica* (Reese, Bozicevich) We have no practical means for making mass stool surveys The complement fixation test, of great value in hepatic amebiasis, is of little or no value in detecting amebiasis limited to the bowel Recently I have found that the liberal use of aureomycin has prevented obtaining a stool specimen for *E. histolytica* in persons who subsequently have been proved to have the disease

Because of these and many other factors it is probably correct to state that we have failed to teach the basic principles of this disease There has been a segmental approach to the problem with the greatest emphasis on public health aspects and laboratory studies of the parasite to the exclusion of emphasis on clinical recognition of the disease This trend is borne out by the fact that a large number of articles on the subject of amebiasis have been written (1500 world-wide, of which 534 had origin in the U S A, from 1933 to 1947); yet D'Antoni pointed out in a review of these articles that few were written on the diagnosis

Even though there are scores of unanswered problems which await clarification in the laboratory, it is important indeed to point out that clinicians interested in amebiasis have greatly increased the number of proved cases

gested. There is not a satisfactory explanation for the mechanism by which the parasite progresses in invasion of an infected host. It is furthermore recognized that large ulcers may exist in the bowel or a large liver abscess may develop without producing symptoms.

The bowel is the primary site of amebic infection, all other tissues invaded by the parasite secondarily from the bowel lesion. Little is known about the body's defense mechanism nor why in one organ a rapid spread occurs while in another the lesion is quiescent. The signs and symptoms of amebiasis may reflect disease in almost any portion of the body.

THE CLINICAL RECOGNITION OF AMEBIASIS

The signs and symptoms of amebiasis are protean and it may mimic many other diseases with generalized or local manifestations Often it is overlooked The clinical masquerade of amebiasis may fall into one of many groups The patient with the systemic manifestations of amebiasis frequently is thought to have tuberculosis, brucellosis, or neoplasm. The intermittently constipated person with vague abdominal pains frequently is thought to have an irritable bowel syndrome. The sudden onset of chills and fever with cough productive of large amounts of chocolate sputum in a patient whose x-ray shows a lung abscess may focus the attention to the chest while the tender enlarged liver with its amebic abscess escapes notice The patient ill for months with vague complaints of indigestion, abdominal discomfort, weight loss, and irritability presents a diagnostic problem. Without careful palpation of the liver amebic hepatitis may go unrecognized The patient with anemia, rectal hemorrhage, anorexia, weight loss and a palpable mass in the abdomen might well be considered as having an incurable carcinoma In the absence of direct proof of neoplasm we must consider amebiasis.

The chance for making a diagnosis of amebiasis lies in suspecting it. The search for ameba must not be confined to adults alone, for contrary to popular opinion D'Antoni (19, 20) has shown that infants and children may be ill with the disease By virtue of the mode of transmission, amebiasis should be considered as a family disease

Boyers (6) listed 1961 separate complaints from 700 patients with amebiasis In spite of this astronomical figure the signs, symptoms, and syndromes can usually be grouped so that they are related to the lesions Thus the clinician must have in mind signs and symptoms initially in the bowel and then along any route of spread to other parts of the body.

The patient's complaints related to the *systemic manifestations* often dominate the picture: malaise, fatigue, an ill-defined feeling of sickness, vague aches and pains with headaches, and weight loss. The patient may have a low grade fever or have periodic chills, fever, and sweats of months'

vaded, resulting in thrombosis and bowel hemorrhage. These infected vessels act as potential foci for seeding parasites to other parts of the body. The cytolytic properties of the parasite enable it to undermine the mucosa The "characteristic" flask-shaped ulcers with a small opening presenting to the lumen of the bowel are an extension of this process. Frequently they are connected by tracts within the bowel walls so that proctoscopic examination reveals small lesions. With more extensive invasion fistulous tracts develop between the peritoneum and bowel. As a general rule the cecal and rectal areas are more likely to be affected though lesions may be found in any portion of the large intestine and rarely the ileum.

Most of the tissues of the body can be invaded by *E. histolytica*. The liver ordinarily bears the brunt of metastatic infections arising from the bowel Knowing that the capillaries and venules of the splanchnic bed are often invaded by the parasites, it is remarkable that serious liver infections are not encountered more frequently. Faust (29) pointed out that the liver parenchyma appears to possess an amebostatic factor On clinical grounds it is suspected that the liver is frequently diseased in the colonic form of the disease Whether this results from products of the ameba, necrotic cells or bacteria is not known. Diffuse hepatitis (10, 18, 56) with invasion of the liver parenchyma results from colonization of ameba in thrombi of small veins in the liver. The amebae spread and cause necrosis of liver cells with little inflammatory response Sometimes such early lesions are reversible if treatment is directed only to the colonic phase of the disease, the source of the infection. On the other hand, with or without treatment areas of necrotic liver tissue may coalesce and produce liver abscesses. Such abscesses range from microscopic size to those occupying the major portion of a liver lobe. The larger ones have a definite wall and are filled with characteristic reddish-brown (anchovy paste) material made up of necrotic liver cells usually free of pus, bacteria, and ameba Often amebae can be found in the abscess wall and adjacent tissues The older the abscess the more likely will be secondary infection Under these circumstances the contents may be purulent.

Amebic invasion of the lung and a resulting lung abscess is rarely blood-born, but usually follows rupture of a hepatic abscess with extension to the lung Brain abscess due to *E histolytica* (46) often is associated with lung or liver amebiasis and the local lesion is comparable to the liver or lung abscess The brain lesion has its origin in the cerebral substance where embolic or thrombotic occlusions of small vessels result in neurolysis. Engman (27) described amebic infections of the skin and they are similar to the lesions of other organs with evidence of marked necrosis of the tissue.

With present knowledge of the pathogenesis of amebiasis it is impossible to predict who will become ill once cysts of *E. histolytica* have been in-

the cecum or rectosigmoid areas. It is hard to distinguish from carcinoma. Ordinarily it is discovered by feeling a mass through the rectum or abdominal wall or incident to an x-ray study of the colon. When this type of lesion is encountered the clinician is obligated to examine the stool for parasites or seek the aid of a competent laboratory. Pericolic abscess and fistulous tracts may be encountered in acutely ill patients and can be mistaken for other granulomatous lesions or neoplastic disease.

Amebic appendicitis occasionally is encountered. The signs and symptoms may mimic those of an acute suppurative or chronic appendicitis. The onset may be sudden yet ordinarily it is preceded by mild cramping pains along the colon, episodes of diarrhea, and change in bowel habits. These minor symptoms may be overshadowed by the more acute discomfort in the right lower quadrant. DeBakey and Ochsner have indicated that 10 per cent of their cases of chronic appendicitis were associated with an amebic infection. If *E. histolytica* is found in the stool of a patient not presenting the classical signs of acute suppurative appendicitis anti-amebal therapy is indicated before exploratory operation.

Frequently the syndromes due to *hepatic amebiasis* are mistaken for other diseases because the signs and symptoms of hepatic amebiasis may make their first clinical appearance in the form of isolated disease in liver, lung, pleura, peritoneum, diaphragm and shoulder or gallbladder. Finally, it may masquerade as a non-localized infection. Under the best of circumstances DeBakey and Ochsner (21, 22) were able to find evidence of amebae or cysts in only 45 per cent of the stools examined, and Banker (3) found in 66 cases of amebic abscess of liver observed at autopsy that 23 per cent had no demonstrable intestinal lesions. Of the cases of amebiasis studied by DeBakey and Ochsner in the past 20 years 11.1 per cent had hepatic involvement. In a collected series of 5,250 autopsy cases of amebiasis these authors found an incidence of hepatic involvement ranging from 7.6 to 84.4 per cent with an average of 36.6 per cent. If liver tenderness be taken as evidence of hepatic involvement in clinical amebiasis, Hamilton and Zavala (33) believe that the incidence is high. Mortality figures for hepatic amebiasis are difficult to interpret because of variables such as virulence of the strain of ameba, type of treatment, economic status, nutrition and quality of medical care. The fatality rate of amebic abscess of the liver varies with treatment and requires comment. DeBakey *et al.* reported an over-all mortality of 22.2 per cent true abscess formation. A 40 per cent mortality was reported when secondary bacterial infection was present whereas in the sterile abscess due to ameba alone it was 5.5 per cent. The death rate from recognized amebic hepatitis without formation of suppuration is nearly zero. On the other hand, an amebic abscess of the liver is a late event, a sequel to amebic hepatitis.

duration without an obvious local infection. Personality changes often accompany the systemic manifestations. There is increased irritability, insomnia, poor memory, and depression which sometimes assumes the proportions of a major mood disorder. These personality changes are certainly a part of the disease for they promptly disappear with proper anti-amebic therapy. Because the systemic manifestations usually have an insidious onset, the patient consults the physician months later. Often one can elicit a story of some irregularities in bowel habits which had appeared to be of no significance to the patient. The physical examination frequently reveals an unexpected degree of liver tenderness or a low grade amebic hepatitis.

Symptoms and signs due to *disease of the bowel* are common and usually are present at some time or other in the course of this disorder. On the other hand, it is an important clinical fact that a serious metastatic complication can develop in the complete absence of enteric symptoms The bowel symptoms are variable in severity and usually are intermittent in character. The acute fulminating dysentery with frequent bloody mucoid stools, tenesmus, great prostration and even deterioration is seen often in tropical countries but rarely in the United States In patients with dysentery one must search for ameba as well as for pathogenic bacteria. Bacterial invaders are in part responsible for the rapid decline and death in acutely ill cases (16).

The usual bowel disorders are characterized by a gradual change in bowel habits with increasing constipation of days' or weeks' duration, alternating with diarrhea. The diarrhea is sometimes explosive but more likely the patient will have a few loose bowel motions for a few days He may have observed blood and mucus in the stool. A few patients will seek aid because of newly developed hemorrhoids.

The patient with colonic amebiasis frequently has abdominal pain and discomfort. Sometimes it is a dull ache over the course of the colon. The pain may occur in the absence of diarrhea or constipation. Portions of the colon often are found to be tender to palpation Bloating is a common complaint and may be associated with nausea and vomiting. Usually there is some degree of griping pain with the diarrhea or loose bowels Often the patient with colonic amebiasis is treated for "idiopathic" ulcerative colitis.

Severe rectal hemorrhage should prompt a search for ameba. Profound rectal hemorrhage was the cause of admission to the hospital in 3 of 32 patients with amebiasis (33) Chronic bleeding from the eroded bowel may be suspected by finding occult blood in a stool specimen. The pallor and anemia in some patients is due to chronic blood loss and in such individuals a story of abnormal bowel function usually can be elicited

Amebic granuloma may form anywhere in the large bowel, usually in

the demonstration of liver tenderness. The ordinary method of liver palpation may fail to elicit tenderness if hepatomegaly is not found. With the following technique of examination Hamilton and Zavala have been able to find hepatitis not otherwise demonstrable: "The examiner stands to the right of the patient and puts his right hand under the right costal margin with the fingertips firmly resting about one-half inch from the xiphoid process. As the patient breathes freely through the mouth the hand is slowly pressed inward and upward. This maneuver presses the falciform ligament of the liver and usually produces pain even though the liver edge is not palpated. If pain is not elicited have the patient take a deep breath. The impingement of the liver on the palpating fingertips produces a sharp pain which abruptly halts respiration. As a control one should palpate in a similar manner in the left epigastrium applying even greater pressure. No pain should be elicited on the left side." Liver tenderness can be elicited in normal persons by deep palpation of the liver, by percussion of the lower chest wall and by marked compression and sudden release of the lateral costal margins Thus the examiner must palpate firmly yet gently and later show that the tenderness disappears with treatment. The patient will be most impressed with the disappearance of liver tenderness and the return of sense of well-being.

D'Antoni (19, 20) has called attention to the fact that often the so-called "normally enlarged liver" observed in children under the age of 6 years was in reality hepatomegaly associated with amebiasis. Reduction in liver size followed in 5 to 10 days under chloroquine treatment. He recommended that all children with enlarged livers should be tested for ameba In young children the sigmoidoscopic examination was the most accurate means of establishing a diagnosis For some unexplained reason children did not pass cysts in their stools and therefore some 80 per cent would have been missed without a proctoscopic examination. Awareness that liver tenderness and vague systemic complaints can be caused by amebic infection will net a real increase in the diagnosis of amebiasis

Severe amebic hepatitis often is associated with clinically active colitis, though it must be emphasized that this is not necessarily true. The patient with severe amebic hepatitis has symptoms of low grade hepatitis with one or more other complaints and signs Frequently there is discomfort or actual pain in the right upper quadrant of the abdomen aggravated by bending forward, jarring of the body while riding in an automobile, or walking The patient may find it through palpation or percussion The pain may be dull aching or sharp and colicky in character The patient may have fevers with drenching sweats and even chills. As a rule a low grade fever is found

Personality changes have been noted in children and in adults. My own

It is impossible to know how long an individual patient may have had an active hepatic amebiasis before symptoms bring him to the physician. It has been relatively common even in small series of cases to find patients who had no significant symptoms prior to the onset of acute symptoms and signs of rupture or impending rupture of a large abscess. Also, it is common to discover an amebic hepatitis in a person who has obviously been ill for many months and who has not developed an abscess Thus, the onset may be acute or insidious, there may be minimal or massive involvement of the liver with little relation to the severity of the symptoms and finally it is not always possible to differentiate amebic hepatitis from amebic abscess on clinical grounds

From the standpoint of the clinical recognition of hepatic amebiasis three forms can be roughly defined: a mild hepatitis, severe hepatitis, and true liver abscess.

Mild hepatitis usually accompanies active amebic colitis. It is characterized by minimal tenderness of the liver which may or may not be enlarged Usually there is no elevation of the temperature or the white blood count. Few if any constitutional symptoms occur and with active bowel disease it is difficult to determine which organs are responsible for the systemic disturbances Sodeman has found ameba by liver biopsy in such situations. This study should dispel any question of the existence of a mild hepatitis Clinical improvement after treatment also has a strong bearing on this point. Treatment of colonic amebiasis with drugs which act primarily in the bowel and cure the colonic phase of the disease relieves the accompanying liver tenderness and hepatomegaly. Also treatment of similar patients with chloroquine directed at treatment of the hepatic amebiasis effects a rapid relief of tenderness but often doesn't cure the bowel disease. A reasonable interpretation of these findings suggests that amebae do cause the low grade hepatitis but do not colonize widely in the liver because of the amebostatic capacity of liver tissue.

There are patients in this group who will be diagnosed only if care is given to palpation of the liver They have tender livers with or without hepatomegaly and often do not have symptomatic bowel disease In this group vague symptoms usually bring a patient to the doctor They are fatigue, ill-defined discomfort with aches, lassitude, loss of strength, depression and irritability. In a surprisingly high number of such patients a careful search of the stool reveals ameba, or the complement fixation test will be positive Treatment of this group with chloroquine causes a remarkable recovery from both signs and symptoms There is even a smaller group who fit this description but do not have laboratory evidence of amebiasis. It is suspected that some patients of this group recover spontaneously

With this history the clinical recognition of hepatitis is dependent upon

response to therapy with chloroquine or similar drugs is presumptive evidence of amebiasis. Patients presenting the characteristic clinical picture must be treated even with negative laboratory tests, particularly if no other cause has been established. Acute hepatitis due to amebiasis may progress to abscess formation. Hepatitis is reversible with medical management while abscess often is not.

The symptoms of *amebic abscess* of the liver may be those of suppuration with chills, fever, sweats, right upper quandrant pain, leukocytosis and demonstrable, painful, and enlarged liver. The clinician's problem in such instances is to consider amebiasis.

On the other hand, amebic abscess of the liver may be camouflaged by the fulminating onset of an overwhelming infection, or attention be distracted by significant signs in neighboring organs Because of the absence of enteric symptoms the true cause of an amebic abscess of the liver is often overlooked.

The patient with amebic abscess usually seeks help after a long illness with fatigue, weight loss, right upper quadrant pain, and one or more of the other complaints described in amebic hepatitis. The patient ordinarily seeks medical aid after a relatively sudden increase in the symptoms, such as the occurrence of chills, drenching sweats, and fevers; symptoms which indicate a progression of hepatitis to abscess. With abscess formation the patient has increased pain in the area of the liver. It may be dull, continuous, or of the intermittent sharp, stabbing type It may be referred to the shoulder if the abscess is in the dome of the right lobe of the liver with irritation of the adjacent diaphragm. With walling off of the abscess the acute symptoms may lessen. But secondary bacterial invasion or loss of local resistance to the ameba aggravates the symptoms. An abscess may become very large before even minimal symptoms such as pain and fever are noted. The sudden onset of chills, fever, progression of the abscess, and rupture into organs of the thorax or abdomen can take place in a few days without any forewarning illness Or the abscess, developed insidiously, becomes walled off but produces no significant local signs. It remains the source of a mild systemic reaction with chronic low grade fever. Jaundice may occur with amebic abscess. The leukocyte count usually is elevated with amebic abscess, the average count being about 20,000. A significant anemia often accompanies the formation of an abscess.

Roentgenography is of great value in the diagnosis of hepatic amebiasis. Positive signs are found in most cases. [illegible]

reinforce the suspicion of hepatic disease. The discovery of a localized upward bulge of the diaphragm is very sound evidence of an abscess. The

experience agrees with that of others that these changes are likely to be present in association with hepatitis. In adults one may observe increased irritability, inability to concentrate, and sometimes manifestations of agitation and depression In children there may be the appearance of the *problem child*, restlessness, and feeding difficulties These personality changes clear rapidly with anti-amebal therapy.

Peculiar skin changes have been observed by Loeber and D'Antoni in children with hepatitis, which they have likened to a "fading suntan". Similar changes are observed in adults and it is my opinion they suggest amebic hepatitis of considerable duration. A description of these changes will be given in greater detail later.

Jaundice is rare in amebic hepatitis and if present is more in favor of an accompanying amebic abscess.

The physical examination usually reveals an easily palpable, enlarged, and tender liver. It is both needless and dangerous to attempt to show further tenderness by percussion of the anterior chest wall. Rupture of an abscess is always a possibility.

The white blood count often is normal. High counts with an increase in polymorphonuclear leukocytes are encountered. Values above 15,000 suggest an abscess or secondary infection yet this is not a steadfast rule. A moderate eosinophilia will be observed in a small number of patients. In our experience in well nourished persons with hepatitis the erythrocyte count and hemoglobin levels are within the normal range. If these values are below normal a more serious complication with pus formation should be considered.

The radiograph of the chest may show an elevated diaphragm and by fluoroscopy it is often restricted in excursion or immobilized. These are valuable findings in confirming the diagnosis of amebic hepatitis.

The symptoms and signs have to be differentiated from those of infectious hepatitis or homologous serum hepatitis The liver damage caused by amebiasis almost always is slight Zavala and Hamilton have studied a patient who had severe amebic hepatitis and abscess and a battery of liver function tests disclosed only minimal abnormality even at the height of the illness. In the recovery phase the functions promptly returned to normal Shute (55) found some change in liver function tests in 50 per cent of 73 cases of amebiasis In viral hepatitis profound alterations in liver function tests are of much greater magnitude than those found in persons equally sick with amebic hepatitis, an excellent point in differential diagnosis.

The diagnosis of amebic hepatitis is established by finding the parasites in the stool. It is highly probable if the complement fixation test is positive. With a negative complement fixation test and negative stool, a prompt

empyema, lung abscess, or hepatobronchial fistula causes an illness of sudden or insidious onset with variable chills, fever, sweats, pain, cough, and sputum The various physical and x-ray findings of fluid, lung consolidation, or abscess are found. None of these is specific for amebiasis. Certain features of each form of pleural pulmonary disease should make one think of amebiasis. The sputum from an amebic abscess or fistula usually has the characteristic reddish-brown color with little or no odor. The pus is relatively free of bacteria and occasionally an ameba may be present. The bacteriologically sterile pleural fluid with or without a chocolate color should suggest an amebic origin When a tender enlarged liver is found by careful attention to the relatively asymptomatic area below the diaphragm, an amebic infection becomes highly probable. The communication of the bronchus with the abscess cavity in the liver can sometimes be demonstrated by the instillation of iodized oil through the trachea.

Rupture of the amebic abscess into the *pericardium* usually causes collapse and death. With slower invasion a benign pericarditis is possible. The pericardial involvement, if recognized, is amenable to active treatment. The tender liver should suggest amebic infection.

The rupture of a hepatic abscess to the *peritoneum* is a serious complication. If the leak is slow it will be walled off, presenting signs of local peritonitis and later a tender mass If the course is chronic and benign, abscesses, masses, and fistula formation may mimic another granulomatous lesion or neoplasm The rupture of a liver abscess with rapid spread producing a generalized peritonitis is almost always fatal. Recently, however, we had a patient under therapy at the time of rupture who survived such a general peritonitis.

Amebic *brain* abscess is uncommon. In their excellent review Orbison, Reeves, Leeham, and Blumberg (46) point up the need for the clinician to keep this possibility in mind when dealing with obscure brain disease. Out of 83 collected cases and their 5 the diagnosis was not suspected in the majority till after death. But one recovery was recorded. An important clinical fact emerged: namely, there was a strong relationship between lung disease, liver abscess, and brain abscess. In only four instances was this relationship absent. The exact incidence of amebic brain abscess is not known From the Armed Forces Institute of Pathology (Orbison) brain abscess was found in 1 2 per cent of 320 autopsied cases coded as amebiasis and in 4 per cent of those with hepatic involvement

The symptoms of amebic brain abscess were focal or general manifestations of central nervous system disease The onset was acute in 50 per cent of the cases Headache and alterations of mental state were the most common symptoms. High fever was frequent There was suggestive but not conclusive evidence that trauma or surgical manipulation of the diseased

differentiation of amebic abscess from infection of the subphrenic space from ruptured appendix or other cause becomes a practical consideration DeBakey and Ochsner have reemphasized the following point: "Most pyrogenic infections of the subphrenic space result from an infection in the appendix and are located in the right posterosuperior space Amebic infections, on the other hand, are located generally in the right lobe of the liver and near the dome, somewhat more anteriorly than posteriorly." Thus anteroposterior views of the chest often show elevation of mid- and medial portion of right diaphragm with a tendency for obliteration of the cardiophrenic angle in amebic abscess In the right lateral view the anterior portion of the diaphragm is elevated with tendency toward obliteration of the anterocostophrenic angle. The pyogenic subphrenic abscess elevates the diaphragm posteriorly and laterally. Left lobe abscess is not frequent It is more difficult to localize. A barium swallow may show pressure deformity with displacement of the stomach and duodenum downward. On rare occasions the use of pneumoperitoneum may be justified to define the relationship of an abscess to the diaphragm This may be an important diagnostic procedure when there has been extension of the process to the pleura and lung.

If a positive diagnosis of amebiasis has not been made and if there is doubt as to the cause of an abscess, aspiration is recommended providing echinococcus cyst is unlikely. The diagnosis of amebiasis is nearly certain if the hepatic abscess contains the reddish-brown or chocolate colored necrotic tissue which is characteristic of this infection The aspiration should be made after two requirements are satisfied First, the patient must be treated with an anti-amebal drug which acts in the liver, and secondly, the aspiration must be carried out under sterile conditions of an operating room A sterile hepatic amebic abscess converted to a bacterially-infected one increases the mortality rate enormously. The use of anti-amebal drugs as a diagnostic-therapeutic test is not reliable for the symptoms may persist until surgical evacuation of the necrotic tissue. If a patient fails to recover promptly with drug therapy the clinician must suspect an abscess.

The invasion of structures within the *thorax* is secondary to an amebic abscess of the liver. The symptoms and signs of the intrathoracic disease may be so prominent or dramatic that they overshadow the underlying liver disease. With invasion or rupture into the right pleura all the signs and symptoms and radiological findings of empyema may be produced. The extension from the liver may be by direct invasion through diaphragm and adherent pleura to the lung producing an amebic pneumonia and lung abscess. The direct extension of the liver abscess to the lung and bronchus may cause a hepatobronchial fistula productive of large volumes of reddish-brown sputum Thus the occurrence of right pleural effusion,

plexion". Locber and D'Antoni (40) compared the changes to a "fading suntan" of the face and pointed out that it is cause enough to suspect amebic hepatitis in children. We believe that pigmentation of the face and forehead is similar to the cloasma of pregnancy. In the South Pacific I observed unusual darkening of the skin of soldiers infected with ameba in contrast to the gray pallor of non-anemic soldiers with malaria. Observations in the temperate climate have revealed similar changes in several adult patients with proved chronic hepatic amebiasis. The color changes in the skin are probably due to an increased melanin Skin biopsy in our laboratory revealed a slight increase in melanin in the basal cell layer but none in the chromatophores. No iron pigment was found and melanin excretion was normal (32) The cause of the pigmentation is unknown. It is associated with hepatic amebiasis of long standing and probably is similar to melanosis noted in other conditions of altered liver or adrenal function The pigment is distributed unevenly over the face, exposed parts, pressure points, in the folds of the skin of the abdomen and flexor surfaces of the wrists as well as over large areas of abdomen and flanks The pigment is accentuated by a fluorescent lamp It disappears slowly with recovery from the amebiasis.

LABORATORY DIAGNOSIS OF AMEBIASIS

Stool and Tissue Analysis

The ultimate proof of an amebic infection lies in the demonstration of the parasite. We must make every attempt to find the parasite, particularly in a disorder which mimics so many other conditions. To increase the number of positive diagnoses the clinician's first responsibility is to obtain an adequate specimen for laboratory examination. Unfortunately the details cannot be left to the imagination of nurse or orderly, and it is at this point that the breakdown in liaison between the patient and laboratory occurs

If the clinician can find in the stool specimen trophozoites which are actively motile, exhibiting progressive motion in one direction, and containing ingested red blood cells, he may make a diagnosis of amebiasis due to *E histolytica* Amebae in this stage are found only in diarrheal and liquid stools or in material obtained directly from amebic lesions Before obtaining the stool specimen the following points should be considered: Trophozoites rapidly die outside of the body A barium enema or barium swallow for x-ray purposes or bismuth used to combat diarrhea temporarily eliminate trophozoites from the feces Urine and mineral oil in the stool render the specimen unfit for examination The clinician must see that a specimen potentially containing motile trophozoites is kept at body tem-

lung or liver favored the complication of the cerebral abscess. The cerebral abscess was brown to port wine or green to yellow in color. Amebae were recovered from the cerebral abscess in 40 per cent of these cases.

Unexplained cerebral symptoms of recent origin call for a careful search for liver or lung disease If it is found ameba must be considered as a common causative factor.

The relationship of amebiasis to *arthritis* has been considered by Perry (47), Rappaport et al (53), Zinneman (64) and others Migrating arthritis, arthralgias, muscle spasm, and arthritis clinically indistinguishable from "rheumatoid" arthritis have been found in patients with amebic infection Many were treated unsuccessfully with gold injections, physiotherapy, sulfonamide drugs, antihistaminics, and aspirin. Rapid recovery from the acute signs and symptoms of arthritis during anti-amebic therapy provided a convincing point for amebic arthritis Some took several weeks for complete recovery. Such amebic arthritis has no specific characteristics It is usually a migrating polyarthritis though it may involve just one joint. Painful swelling of the fingers, knees and ankles may be present. The fingers may have the fusiform deformity of rheumatoid arthritis. The shoulders, neck and back are common sites of pain and muscle spasm. Urticaria occurred in two of four cases of amebic arthritis reported by Rappaport. A prompt response to treatment for amebiasis is expected. In most of the reported cases the colonic or hepatic symptoms of amebiasis were minimal and often were discovered after unsuccessful attempts to treat the arthritis The mechanism for production of amebic arthritis is unknown Amebae have not been found in the joint

The direct invasion of the *skin* by ameba was called amebiasis cutis by Engman and Heithaus in 1919 (26). A review of the clinical and pathological status was made by Engman and Meleney in 1931 (27) The lesions of amebiasis cutis *are irregular ulcers of the skin with overhanging edges of* drying epidermis and a floor of granulations, pus, and debris The margins often have a dusky-red color. Ameba can be demonstrated in the margins The amebic ulcerations are almost always secondary to a draining sinus or amebic colitis. They may develop on the skin at the site of drainage of a liver abscess, a drained appendiceal abscess, or from enteric cutaneous fistula *Ulcerations* may be found about the buttocks, perineum or anus in association with amebic colitis. In rare instances no connection with a visceral lesion can be established

Hyperpigmentation of the skin is another dermatological manifestation of amebiasis This poorly defined change in the skin has been described variously as "sallowness of the skin", "copper penny color", "bronze pigmentation", "subicteric look", "doughy inelastic myxoid complexion", "anemic appearance", "slight jaundice", and "peculiar muddy com-

growth. This important discovery will enhance the study of the growth factors and effect of drugs on ameba (49, 50). If direct smear and concentration methods fail to demonstrate parasites, cultural and staining methods should be employed. Laird, Drinnon, and Davis (39) have shown in a comparative study the advantage of cultural methods Of one group of 350 subjects studied 48 positive results were obtained by culture but only 28 by staining of fecal smears A comparison of specimens obtained from spontaneous bowel movements and ones obtained directly through the proctosigmoidoscope have clearly shown that specimens may commonly be positive by one or the other method only. As has been previously mentioned this is particularly true in children. Thus to increase the number of positive diagnoses the direct collection of material from the bowel is recommended as a valuable procedure and should be included in the routine examination for parasites.

The two-vial method permits the collection of fresh liquid or mushy and solid stools and their shipment by mail to a distant laboratory where reliable examinations can be made by highly skilled workers. The first vial contains polyvinyl alcohol for preservation of liquid specimens The second vial contains 10 per cent formalin for preservation of specimens of solid stool In 1948–49 Goldman introduced polyvinyl alcohol (P.V.A.), a fixative and preserving agent for study of active motile or vegetative forms of ameba, ciliates and flagellates found in liquid stools This preservative [illegible] n [illegible] g solutions At the laboratory, smears can be prepared and stained immediately, or smears can be made and stained months later. The ratio of fresh stool to P.V.A must be exactly 1:2. The formed stool is put in the vial containing 10 per cent formalin in a ratio of 1 part stool to 9 parts formalin for the detection of cysts of ameba and ova of other parasites. It is suitable for analysis by concentration methods.

The efficiency of different methods of stool examination has been compared (30, 37, 59, 60, 62) From these studies it is clear that one single test to the exclusion of others is not efficient in detecting ameba Many tests must be employed and must be repeated over and over again to get maximal results. The zinc sulfate floatation concentration technique is around 70 per cent efficient in finding cysts in persons *known to harbor E. histolytica* and 100 per cent efficient by the fifth examination (59, 60) These authors demonstrated that it is more effective to repeat stool analysis on a second stool rather than on the first specimen It has been known that the concentration of cysts varies enormously from specimen to specimen in an individual The zinc sulfate floatation method is applicable only to cyst

perature and examined promptly! If diarrhea is not present and a search is to be made for trophozoites a liquid stool must be produced by purgation This is best accomplished by giving the fasting patient 3 glasses of water to be followed in a few minutes with 2 ounces of a saturated solution of sodium sulfate (magnesium sulfate has a greater tendency to slow the motility of the ameba). The liquid stools are collected individually in numbered cardboard containers and are examined immediately. Delay beyond 30 minutes reduces considerably the number of positive examinations Therefore it is mandatory that the laboratory be alerted in advance of the study Even in Korea where acute amebiasis was present in our soldiers and the stools frequently were teeming with parasites Hardy reported that the number of positive stools in patients with bowel disorders was much increased when circumstances moved the laboratory of an Army Hospital into the ward. The need for rapid transfer of specimen to microscope stage cannot be over-emphasized A fleck of mucus or blood-tinged mucus is removed from the warm liquid stool with a platinum loop, transferred to a warm slide, mixed with an equal amount of warm saline and covered with a coverslip The slide is examined on a microscope stage which has been preheated with a portable light bulb or by a microscope stage incubator. One is more likely to find ameba in mucus than in particles of feces

In the event of failure to find the trophozoites of *E. histolytica* other techniques must be used either to demonstrate the cystic stage of the parasite or to grow the organism in culture This requires highly trained technicians and adequate laboratory facilities The cysts of *E histolytica* may be identified in the unstained state or by one of two staining methods generally employed. Fresh concentrated specimens from solid stools can be stained with iodine, preferably D'Antoni's standardized iodine stain. Iron-hematoxylin stains of material either from culture, feces, or tissues make permanent preparations They are invaluable at times in the final differentiation of *E. histolytica* from non-pathogenic ameba It is beyond the scope of this review to describe in detail the techniques and morphology of identification of the parasite under discussion For these details reference should be made to Craig and Faust (18) and Elishewitz (25) with their attending bibliographies

The concentration of cysts by the zinc sulfate floatation method and final staining of the parasite with D'Antoni's iodine is a relatively simple and reliable technique Cultivation of ameba on media, a proved laboratory procedure, has the advantage that cultures may be started from cysts or trophozoites The successful culture of ameba on artificial media requires the presence of bacteria Recently Phillips (48) substituted *Trypanosoma cruzi* for bacteria in the culture medium and found that it supported

patients with malaria, kala-azar, echinococcosis, infectious hepatitis, cirrhosis, and carcinoma of the liver, all disorders which could be mistaken for amebiasis. False positive reactions have occasionally been recorded but usually the reaction is weak

Craig has pointed out the value of the complement fixation test in the control of treatment and feels that a persistently positive test means residual infection Terry and Bozicevich (58) discussed this problem in relationship to hepatic amebiasis. After treatment is begun the test may become negative within two to three weeks If the test remains positive for several weeks retreatment is recommended. A persistent positive complement fixation test after several courses of therapy must be evaluated in light of the clinical findings. If the patient remains well he should be considered free of the disease.

Sigmoidoscopic Examination

This is a valuable aid in the diagnosis of amebiasis. It provides otherwise unobtainable specimens for laboratory study and makes possible examination of ulcers in the bowel which may harbor *E. histolytica*. It permits the study of the healing process of active lesions. Evidence of active disease will be observed in no more than a quarter of all cases studied. From a practical standpoint we have divided the lesions observed into four groups. The first group is composed of superficial discrete red to brown papules 1 to 3 cm. in diameter. The rest of the mucosa appears normal. These papules form small clusters in the rectal ampulla and on the rectal valves in the lower 4 to 5 inches of the bowel. Because they are small and few in number they may be overlooked or attributed to trauma of the cleansing enema. However, it is not uncommon to find the same lesion on repeated examinations even after therapy has been started This lesion is probably as "characteristic" of colonic amebiasis as one can find. These papules (Radke) usually disappear by the tenth day of treatment. Specimens are best secured from these and other bowel lesions with a 1 ml blunt-tipped serologic pipette with an ordinary rubber suction bulb A metal curette can be used. These instruments should be on hand at the time of proctoscopy. Cotton swabs should not be used for procuring specimens because amebae adhere to the fibers and cannot be transferred to the slide. The second group consists of distinct ulcers with ragged overhanging edges measuring from 1 mm to 1 cm or more in diameter. A suspicious point is the normal intervening mucosa. Careful inspection often reveals the pinhead size lesions characteristic of *group one* adjacent to larger ulcers. In group three the ulcers are larger with necrotic bleeding bases with connecting bridges of granular mucosa. The fourth group consists of a granular hyperemic mucosa which bleeds with slight trauma. It is important to note that the lesions

examination for zinc sulfate solutions cause rupture of vegetative forms of ameba. Single slide and stain examinations of fresh stools are probably half as effective as the concentration method. The ideal examination would make use of direct smear of liquid and solid stools, concentration methods, iodine and iron-hematoxylin stains, and analysis of freshly obtained specimens fixed in P.V.A.

The contents of an amebic abscess are often sterile and even free of ameba If the wall of such an abscess is explored a section for laboratory examination should be taken from the tissues adjacent to the viable parenchyma for it is in this zone that amebae are most likely to be found Specimens to be studied in fixed tissue sections must be placed in formalin immediately. Ameba may not be recovered from abscesses or ulcers which harbor this parasite because pus or necrotic debris rather than proper specimens was submitted to the laboratory.

Complement Fixation Test

This serological test has proved to be a valuable adjunct in the diagnosis of amebiasis. The range of its specificity in all forms of amebiasis is unsettled (7) Craig (1927–29) was the first to perfect this test and place it on a sound basis for human amebiasis. Subsequent confirmation leaves little doubt of the specificity of the test, but unfortunately there have been technical difficulties in preparing pure and stable antigens effective with all strains of *E. histolytica.* There is a difference of opinion on the exact methods for doing the test. The more sensitive the test is, the less specific it is False negative and false positive reactions occur. These and many other factors make it impossible to use the test as a routine screening procedure. At the present time with the available information (7, 17, 24, 38) the complement fixation test is likely to be positive in moderate and severe amebic hepatitis and amebic abscess of the liver It is less reliable in the colonic amebiasis now encountered in the United States. The test is frequently positive in hepatic amebiasis even though the ameba cannot be found in the stool. A clinical history suggestive of amebiasis together with a positive complement fixation test constitutes very strong evidence for a diagnosis of amebiasis If the test becomes negative with treatment the diagnosis is practically confirmed. In our experience the complement fixation test in hepatic amebiasis has been exceedingly helpful in obscure cases Exceptions are illustrated by a recent patient with a large amebic liver abscess whose complement fixation test was negative. Amebae were demonstrated in the wall of the abscess on direct examination, by stains, and in tissue sections (Borts). Thus a negative complement fixation test does not exclude amebic abscess

Hussey et al. (38) found negative complement fixation tests on sera from

tients if drainage is required. Furthermore, if drainage is necessary closed aspiration is the desirable procedure

A definite principle concerning the duration of drug therapy cannot be established It depends in part upon the toxicity and specific action of the drug. Radke (51, 52) has suggested the optimal treatment time as 25 days based on the observations that bowel ulcers heal in 15 days and the remaining pigment spots in bowel disappear 10 days later. He also feels that pigment spots may harbor latent infection and cause relapse. A review of many forms of therapy suggests that with the short term 7-day treatment too many failures occur With most of the available non-toxic drugs courses of 10 to 21 days are most desirable.

A final principle recognizes the variability in individual response to a given drug Thus regardless of drugs used "routinely" the clinician may have to change drugs to effect a response.

An evaluation of the comparative worth of the available anti-amebal drugs used in clinical medicine is impossible because of the inherent difficulties of setting up truly controlled clinical experiments. A continued search is being made for better compounds and for more effective use of existing drugs. The physician who sees many cases of amebiasis has set routines of therapy and obtains good results, but even under ideal circumstances a few failures occur.

It is presumptuous to offer the "best" program for the treatment of amebiasis although the physician who has an occasional case of amebiasis to treat is confronted with the very practical question of what drugs to use. I therefore am going to discuss the available drugs of proved value or promise in the treatment of amebiasis and point up certain virtues and disadvantages in each

We have drugs acting chiefly on the metastatic tissue phase and those more effective in the primary lesions of the bowel. At least one drug, then, should be selected from each group and used concurrently. It is my conviction that initially the least toxic drugs should be used in treating uncomplicated cyst passers and patients with amebic colitis or amebic hepatitis.

Treatment of Tissue Phase

Drugs primarily effective on the trophozoite in the tissues remote from the ulcerative lesions in the bowel wall are emetine, chloroquine, quinacrine (atabrine), and sontoquine

EMETINE

Emetine hydrochloride is a time-honored drug which has proved its effectiveness in the treatment of amebiasis, particularly the acute forms

of the last two groups cannot be differentiated from those of "idiopathic ulcerative colitis".

To obtain optimal results the clinician must carefully supervise the preparation of the patient for proctoscopy. Luke-warm tap water enemas are recommended. 0.9 per cent saline enemas are ideal. Soapsuds enemas intercfere with examination, for the parasites are killed and the mucosa becomes injected, obscuring minute ulcerations which might otherwise be observed

TREATMENT

With available drugs a high cure rate can be expected provided attention is given to the principles of the treatment. These principles are based on the pathogenesis of the disease and the pharmacological characteristics of the drugs. The basic problem is to obtain a high concentration at sites of infection. There is no single drug which can attain a high enough concentration in both the bowel and the tissues generally to destroy all the parasites in these different locations As a generalization, drugs which are poorly absorbed by the small bowel exert their major effect on the parasites in the ulcers and tissues of the bowel wall but have little or no effect on ameba deep in the tissues, splanchnic capillaries or distant from the bowel On the other hand, anti-amebal drugs absorbed rapidly in the circulation kill parasites in the tissues but fail to eliminate ameba from the ulcers and abscesses adjacent to the gut From earlier discussion it follows that once a diagnosis of amebiasis is established the clinician must assume that both the tissue and bowel phase exist Thus to be assured of a rapid recovery and a low relapse rate at least two drugs must be given: one, a poorly absorbed anti-amebal drug which will localize in the bowel, and another which is rapidly absorbed and reaches high tissue concentrations. It follows also that these drugs must be given concurrently to obtain the optimal effect.

A second principle of therapy recognizes the concept that bacterial infection either in the ulcers of the bowel wall or the abscesses of metastatic lesions interferes with rapid recovery from amebiasis Ulcers of the colon are usually quite free of bacteria when active amebae are present. Under anti-amebal therapy bacteria replace the disappearing amebae. Thus an appropriate drug should be directed to the treatment of secondary bacterial invaders. In the more severe amebic infections antibiotic drugs should be used as an adjunct to anti-amebal treatment

A third principle concerns the therapy of amebic abscess. As a general rule such abscesses should not be drained surgically until adequate medical treatment has been initiated Many amebic abscesses heal under drug treatment. Also it is recognized that survival rates are higher in pretreated pa-

reactions are to be expected. The clinician has the responsibility for stopping the drug where such symptoms appear. For a review of the toxic manifestations the reader is referred to the paper by Klatskin and Friedman (38a)

CHLOROQUINE

The addition of chloroquine to the list of anti-amebic drugs by Conan (1948) was an outstanding contribution because for the first time we have a relatively non-toxic drug about as effective as emetine in the treatment of hepatic amebiasis. Chloroquine is a member of the 4-aminoquinoline series and has been used extensively in the suppression and treatment of malaria It has not produced serious toxic reactions. During the course of treatment mild headache, minimal visual disturbances, pruritis and slight gastrointestinal upsets have been encountered occasionally. In man about 90 per cent of the drug is absorbed. Conan in 1950 showed that the concentration of chloroquine in the liver of human volunteers was 1500 times that of the plasma. The reports of Conan, Sodeman, Manson-Bahr, Murgatroyd and others show that chloroquine is an effective drug in the treatment of amebic hepatitis, amebic abscess, draining sinuses, pleural amebiasis and in my experience peritoneal abscess It is not efficient in the bowel phase of the disease presumably because of lower concentrations The relapse rate of persons treated with chloroquine probably is low, though a large series of cases has not been reported. An analysis of relapse rates should take into consideration the use of drugs used to control the intestinal infection. We have been much impressed with the apparent cure rate following chloroquine therapy. In our experience and that of others the symptomatic improvement is usually dramatic. In amebic hepatitis and small abscesses of the liver tenderness disappears in a few days and the organ recedes The response is ordinarily a little slower than with emetine. A number of patients with hepatic amebiasis with and without drainage who have not responded to emetine have made a rapid recovery when given chloroquine Sodeman reported initial failure of the drug in 3 patients but later the drug was effective in controlling the infection. In these cases fever was continuous after therapy was begun with chloroquine but recovery eventually followed surgical drainage This emphasizes the facts that a failure to respond to therapy does not rule out the diagnosis of amebiasis, that the drugs may not be effective in some instances of pus formation, and that with adequate surgical drainage the drug may then help cure the infection. Such considerations must be given when evaluating any of the new amebacidal drugs.

The dose of chloroquine advised by Conan has been satisfactory: priming dose of 1 0 gram of chloroquine diphosphate daily for 2 days and followed by 0 5 gram of chloroquine diphosphate daily for 21 days.

of the disease. It is a protoplasmic poison for both ameba and human tissues. The therapeutic dose is close to the toxic and in some instances the lethal dose. Until recently it has been the drug of choice in non-intestinal amebiasis. Introduction of the non-toxic chloroquine, which is probably equally effective, has reserved emetine for special types of amebic infection. Emetine is specifically indicated in gravely ill patients for it is the most rapidly acting drug. Its effect is apparent in a few hours after injection even in moribund patients It should be administered after accidental surgical incision of unsuspected amebic abscess. We use it in amebic abscess or complicated amebic lesions which have failed to respond to other drugs Even though emetine is not curative in bowel lesions it has a dramatic effect in some instances of amebic dysentery refractory to other forms of treatment. It has been an excellent drug for the treatment of hepatic amebiasis and should serve as a standard to compare the effectiveness of newer compounds used in extraintestinal amebiasis.

Because of the very real possibility of severe toxic symptoms and even death the total dosage, method of administration, and clinical care must be supervised carefully. Usually it should not be given to children. It is best tolerated by subcutaneous injection Intravenous administration is dangerous. Intramuscular injection may produce local muscle spasm, hemorrhage, and a local weakness which will persist for several days In adults the daily dose should not exceed 0.065 grams (1 grain) and it should be given for not more than 7 days with a total dose of 0.455 grams (7 grains). Because of the cumulative effect and slow excretion a course should not be repeated for a month Patients under emetine therapy should be kept in bed. Some undesirable and toxic symptoms occur in most patients treated with emetine There is usually a marked hypotension the second or third day. Varying degrees of asthenia, true muscle weakness, and fatigue are to be expected and should not be interpreted as results of the disease Sudden shifts in position with syncope or calls for strong muscle activity may endanger the patient's life Because of these toxic symptoms the patient should not be treated on an ambulatory basis. During the course of treatment emetine may produce diarrhea simulating a recurrence of the dysenteric symptoms Nausea and vomiting are thought to be central in origin Electrocardiographic tracings should be taken before therapy is started and perhaps on the third and sixth day of treatment and two weeks after it is completed. If the disease is not of a critical nature any significant changes in the P-R interval or T-wave call for the withdrawal of the drug for myocardial damage may occur and be progressive Dach and Moloshok have shown that the EKG changes may develop up to two weeks after the drug is discontinued It is suggested that with progression of any of the toxic symptoms the drug be stopped In doses recommended few serious toxic

are effective against ameba on the intestinal mucosa and those at some depth in the amebic lesion These compounds maintain high concentration in the large bowel which accounts in part for their effectiveness in the colon They are absorbed to varying degrees (35) but not to an extent they are reliable in the treatment of extraintestinal lesions. They are practically non-toxic drugs and have relatively high anti-amebal qualities From published reports it is impossible to determine with any degree of certainty which is best

Diodoquine has the lowest incidence of side reactions and is ideal for use in ambulatory patients as well as those suffering with the most severe forms of the disease. On occasion a mild headache has been reported or an unusual taste in the mouth. The drug is given in 630 mg. tablets 3 times a day (total daily dose of 1890 mg) for 21 days Diodoquine has been recommended in the therapeutic regime by the U. S. Army When used concurrently with chloroquine or emetine for example, a cure rate of 85 to to 95 per cent can be expected.

Chiniofon (yatren, anayodin) was the first of these drugs to be used in amebiasis (1921) and has been highly effective in the treatment of cyst passers or "carriers" as well as in the more acute form of the disease It has the same range of usefulness as diodoquine but in some instances produces severe diarrhea. The mild diarrhea that sometimes develops usually clears up while therapy is continued. The adult dose is 0 25 gram (4 grains) 4 times a day for 10 days.

Vioform may produce abdominal cramps, nausea, vomiting, diarrhea, and dyspnea though ordinarily little or no discomfort is noted. It is recommended that 0 25 gram of the drug (in gelatin capsule) be given 3 times a day for 10 days with a rest period of one week followed by another 10-day course.

Carbarsone, thiocarbarsone and *milibis* are three of several arsenicals which have been successfully used in the treatment of intestinal amebiasis. Other arsenical drugs will not be considered for they are no more effective and are more toxic.

Carbarsone. An evaluation of the therapeutic effect of carbarsone is particularly difficult for 90 per cent cure rates to 100 per cent relapse rates are reported Radke observed ulcerative relapse occurred in 3 patients during the time adequate carbarsone was being given Carbarsone has been fairly effective in elimination of cysts from carriers and mildly ill patients The drug is relatively safe if given in doses recommended though the usual dangers of arsenical poisoning are to be considered The drug should not be given to people with kidney or non-amebic liver disease Previous use of arsenicals might also be a contraindication Carbarsone is given 0 25 gram (4 grains) twice daily for 10 days and may be repeated in 10 days. Mild

QUINACRINE (ATABRINE)

In a search for safe and effective anti-amebic drugs Radke (51, 52) believed the significantly lower incidence of *E. histolytica* in the stools of soldiers returning from the Pacific as compared to those from Europe might be due in part to the widespread use of atabrine. He therefore used atabrine alone and in combination with carbarsone and/or aureomycin in the treatment of amebiasis. The results of these excellent studies show that quinacrine alone may produce a rapid remission and cure in subjects with acute amebic dysentery and amebic hepatitis. When combined with carbarsone and aureomycin in cases of hepatic abscess and pleuropulmonary involvement there was recovery. Failures were reported in the quinacrine-carbarsone-aureomycin treated groups. It appears that quinacrine is effective in treating both the tissue and bowel phase of the disease. Further studies are needed to evaluate the relapse rate after use of this drug, compared to other drugs. While in the Pacific I observed proved severe amebiasis progress in soldiers being treated with massive doses of atabrine for malaria. Recovery from the amebic infection was brought about only after combined emetine, chinofon, and carbarsone treatment Atabrine is reserved for treatment when other drugs have failed The dosage recommended is 0 1 gram 4 times daily for 15 days

SONTOQUINE NAPHTHOATE

Another 4-aminoquinoline derivative has recently been used by Conan (14) in an attempt to find a single drug effective against amebic hepatitis and intestinal amebiasis *About 80 per cent is absorbed compared* with 90 per cent of the oral dose of chloroquine Sontoquine naphthoate was effective in hepatitis and controlled symptoms in intestinal amebiasis though in the latter group half the cases treated continued to pass *E. histolytica* in the stools. Qualitatively it appeared to have activity similar to chloroquine

Treatment of Bowel Phase

The following drugs are primarily effective in the colonic phase of amebic infection: diodoquine, vioform, chiniofon, carbarsone, thiocarbarsone, bismuth glycolylarsanilate (milibis), aureomycin, and terramycin. Other drugs [illegible] ...al This list has been [illegible] trial and are readily [illegible] een too readily available. Such a long list of drugs makes it very clear that we do not yet possess the ideal amebacidal drug

Diodoquine, vioform, and chiniofon are iodoquinoline compounds which

aureomycin. Further evaluation of this drug is needed, particularly with reference to dosage and duration of therapy.

Terramycin has amebacidal qualities in relatively asymptomatic as well as in severe amebic dysentery when the clinical response was as satisfactory as with emetine (presumed to be control of acute symptoms) as reported by Most and Assendelft (44). Seven patients with acute colonic amebiasis with bloody diarrhea, abdominal cramps, and six with right upper quadrant pain were treated for 14 days with large doses of terramycin by Killaugh and Magill The acute symptoms disappeared promptly and the stools became negative for ameba The hepatitis responded more slowly. In one case the signs and symptoms of a hepatic abscess developed while the patient was under treatment with terramycin The acute symptoms of the abscess subsided under chloroquine treatment alone, and complete recovery followed aspiration of the abscess. Four of the remaining cases were followed adequately. Two of these had a recurrence of ameba in the stools. These results show that terramycin cannot be relied upon for control and cure of acute amebiasis Tobie, Most, Reardon and Bozicevich (59) studied this drug in asymptomatic amebiasis The prevalence rate of 222 persons was 49 per cent before treatment After treatment there was virtual elimination of the infection even at the end of the follow-up period of six months The dose used was 2 grams daily for 10 days in patients over 75 pounds. These studies are encouraging in the control of the asymptomatic phase of the disease in large groups.

Both terramycin and aureomycin should be administered for 10 to 15 days in 2 to 3 gram doses daily to obtain optimal results. Their use should be directed to the bowel phase of the disease and to secondary bacterial invaders. In clinically active disease the drugs should not be used alone but in conjunction with other anti-amebal drugs.

Bacitracin, a poorly absorbed drug, has been effective in intestinal amebiasis. Most, Miller, Grossman, and Conan (43) found an approximate cure rate of 33 per cent In one case bacitracin produced a remission of a severely ill patient who had failed to respond to chloroquine, diodoquine, emetine, and milibis The recommended dose is 80,000 units daily for 10 days. In the case reported above 160,000 units were administered daily.

From the standpoint of therapy the ill patients can be divided into three groups There are those with mild to moderate disease of the colon and liver. They can be treated on an ambulatory basis and respond well to combined therapy with chloroquine and diodoquine given for 21 days. Results of treatment will be good if any of the other iodoquinolinol or arsenicals are substituted for diodoquine. In clinically active disease of this sort it is the author's opinion that terramycin and aureomycin should be used only in addition to some other drug effective on the bowel phase of the infection.

gastro-intestinal upsets are encountered. It is my opinion that this drug should be reserved for cases resistant to diodoquine or chiniofon.

Thiocarbarsone, a thioarsenite related to carbarsone oxide, is much less toxic in animal experiments than carbarsone. A cure rate of 90 per cent was reported by Anderson in 1949. In some patients with drug refractory amebiasis thioarsenites have been of great benefit. Anderson reported that retention enemas with these compounds cleared ulcers in stubborn cases Because the drug may cause severe nausea and vomiting tablets are enteric coated. The recommended dose for adults is 100 mg. 3 times a day for 10 days. The exact place of this drug in the therapy of amebiasis has not been established. It certainly may help after other drugs fail. One manufacturer of this drug has suggested that it should not be used in amebic hepatitis or in the presence of impaired liver or kidney function which precludes its use in many cases of amebiasis.

Milibis, or bismuth glycolylarsanilate, a relatively insoluble compound, has shown considerable promise in the treatment of intestinal amebiasis. It is of low toxicity though in excessive doses Brown (1950) observed a case of arsenical encephalitis. Berberian, Dennis, and Pipkin (4) presented data indicating that milibis was 90 per cent effective whereas chiniofon was only 47 per cent effective in eliminating ameba from the intestinal tract In this series of experiments it might have been a more convincing comparison had chiniofon been given for a greater length of time. Nonetheless concurrent therapy with chloroquine and milibis will probably prove to be an efficient method of treatment. The adult oral dose of milibis is 0.5 gram 3 times a day for from 7 to 10 days

Aureomycin, terramycin and bacitracin have been used in the treatment of amebiasis with varying degrees of success A complete evaluation of these drugs is dependent upon further clinical trial.

Aureomycin has been used by a number of authors (review by Calero) and reported as ranging from totally unsuccessful to 100 per cent curative From a review of many case histories and our own experience it is apparent that aureomycin will clear the stool of parasites but that relapses occur, in some series in high incidence. Aureomycin cannot be relied upon to control the extraintestinal lesions With further experience increased doses of aureomycin undoubtedly have increased the efficiency in the bowel phase of the disease (36, 41) We have observed a mental patient with rupture of a large amebic abscess producing generalized peritonitis This rupture took place while the patient was taking aureomycin In spite of long treatment with aureomycin trophozoites of *E histolytica* were found in the abscess wall after surgical drainage. Eventual recovery took place during treatment with chloroquine and diodoquine We believe that this patient survived the rupture hitherto fatal because of the coverage with

tening the resolution of acute or chronic amebic abscesses. *In vitro* studies by Sherry, McCarty, and Tillett (54) have shown that an abundance of extracellular desoxyribonucleoprotein was responsible for the viscous coarse qualities of the exudate of amebic abscess of the liver. Such an abscess was treated by aspiration and the instillation of solutions of streptokinase and streptodornase. The instillations were made through two plastic catheters with an inside diameter of 1.8 mm. inserted into the abscess cavity at the time of initial drainage. One served as an inflow and the other as an outflow tract. Instillations were made at 4-hour intervals for 24 hours. A rapid recovery was observed. The drainage became thin and ceased in 4 days. The patient was also treated with chloroquine and terramycin, drugs which probably contributed materially to the rapid recovery once the contents of the abscess cavity had been liquified and removed.

If pleural amebiasis develops conservative management should be followed If indicated, aspiration of the pleural space and underlying abscesses should be carried out under aseptic conditions. The more chronic empyemas, persistent bronchobiliary fistula and lung abscesses are amenable to thoracotomy, decortication of the lung and diaphragm, and lung resection.

If an amebic abscess of the brain is recognized Orbison has recommended the use of antibiotics and anti-amebic therapy (chloroquine and emetine) combined with needle aspiration through an exploratory trephine The initial problem, then, is to maintain life and later neurosurgical procedures will be directed to the management of a chronic brain abscess

POST TREATMENT

After a patient has completed a course of treatment and recovered from the immediate effects of the amebic infection the clinician is faced with the question of cure Is the patient truly free of infection? The judicious evaluation of the clinical course and the laboratory studies over several months will ordinarily answer the problem Nearly every person who has been treated for active amebiasis will express delight in the improvement of a sense of well being and increased physical stamina The patient will "look back" and state, "I didn't realize how badly I felt." In spite of the improvement and recovery a few patients who had colonic amebiasis will experience episodes of vague abdominal cramps and loose bowel movements assuming more the character of an irritable bowel syndrome Sigmoidoscopic examinations and stool studies must be repeated and when negative, reassurance, low residue diet, antispasmodics, and regulation of the bowel habits is usually sufficient treatment. In a matter of several weeks recovery is ordinarily complete The relapsing patient will have an increase in the severity of the symptoms and will often state, "I felt this way when I was getting sick before." A considerable amount of weight should be given to this

The second group are those more acutely ill patients with dysentery or hepatitis who should be confined to bed or a hospital and treated as outlined in the preceding paragraph. In addition an antibiotic should be administered. Rapid and complete cure is anticipated. In the event severe enteric symptoms persist emetine should be given subcutaneously for often it alone will bring about a rapid resolution of the acute symptoms. If treatment fails additions of other drugs from those discussed are recommended. Whatever treatment schedule is elected the drugs must be used in full doses and for the duration recommended. With persistence of symptoms and fever the clinician must seriously consider an amebic abscess

The third group are patients with amebic abscess. DeBakey and Ochsner (21, 22) have made a careful summary of 20 years' experience of the treatment of this more severe form of the disease. An evaluation of the multiple factors in the successful therapy of hepatic amebiasis has been made by Sodeman *et al.* With the tentative diagnosis of amebic abscess of the liver or a collection of amebic "pus" elsewhere the initial therapy is conservative Chloroquine or emetine is given, the latter in the most acute form of the disease, and in addition an appropriate antibiotic. Some abscesses will heal under this regime. Persistence of fever and signs of the abscess are indications to evacuate the pus. If this decision is made early in the illness DeBakey and Ochsner have warned that emetine should be given first and preferably for many days Emetine and antibiotics will help prevent local and/or systemic spread of infection when the drainage is carried out, or if rupture of the abscess should occur

If an amebic abscess becomes secondarily infected the mortality rate is very high In DeBakey's series the mortality was 40 per cent in the bacterially infected group and 5.5 per cent in the sterile (pure amebic) group Thus the closed type of aspiration drainage under sterile conditions in an operating room is the desired surgical procedure Furthermore in the best of hands open drainage brought a mortality rate of 22.2 per cent whereas with the closed aspiration type of drainage it was only 4 0 per cent These authors have also shown that the extraperitoneal routes of drainage materially reduce the mortality rate If it is demonstrated by smear and culture that the abscess is secondarily infected open drainage may be resorted to Usually drainage of an uncomplicated abscess is followed by drop in temperature and rapid recovery Repeated aspirations occasionally are necessary. Drug therapy should be continued during this recovery period and antibiotics continued until drainage has ceased. If an abscess ruptures spontaneously to a bronchus to form a hepatobronchial fistula surgical drainage is ordinarily not necessary but it is best treated with drugs and postural drainage.

Streptokinase-streptodornase enzymatic solutions may be useful in has-

biasis treated with choroquine. Trans Roy. Soc. Trop Med. & Hyg , 43: 659–666, 1950.
14. CONAN, N J , JR : Santoquine naphthoate in amebiasis. Am. J. Trop Med., 31: 18, 1951
15 CRAIG, C. F.: The Parasitic Amoebae of Man. J. B Lippincott Co , Philadelphia & London, 1911
16 CRAIG, C F.: The Etiology, Diagnosis, and Treatment of Amebiasis The Williams & Wilkins Co , Baltimore, 1944.
17. CRAIG, C. F Amebiasis and the complement fixation test. U. S. Armed Forces M. J , 1: 1337–1342, 1950.
18. CRAIG, C F. AND FAUST, E. C : Clinical Parasitology, 5th Edition. Lea and Febiger, Philadelphia, 1951
19. D'ANTONI, J. S The pattern of the literature of amebiasis. Am J Trop. Med , 29: 269–293, 1949.
20. D'ANTONI, J. S.: Amebiasis Panel, Discussion Am J. Trop Med , 30: 144–146, 1950
21. DEBAKEY, M AND OCHSNER, A : Surgical treatment of amebiasis Wisconsin M. J , 48: 243, 1949.
22 DEBAKEY, M. E. AND OCHSNER, A . Hepatic amebiasis A 20 year experience and analysis of 263 cases Surg Gyn. & Ob , 92: 209–231, 1951
23. DENNIS, E. W : Amebiasis Panel, Discussion Am. J. Trop. Med , 30: 159–164, 1950
24 DOLKERT, R E , HALPREN, B. AND CULLEN, B. A : The diagnosis of amebiasis, the role of the complement fixation test, and the incidence of the disease in the Chicago area J. Lab & Clin. Med , 38: 804, 1951,
25 ELISHEWITZ, H.: Recent advances in parasitological diagnostic technics Am J. Med Technol , 17: 165–189, 1951.
26 ENGMAN, M. F , SR AND HEITHAUS, A S : Amebiasis cutis. J Cutan Dis , 37: 715, 1919.
27. ENGMAN, M F., JR AND MELENEY, H. E · Amebiasis cutis (Endameba histolytica) Arch Derm & Syph., 24: 1–21, 1931.
28 FAUST, C Amebiasis in the New Orleans population as revealed by autopsy examination of accident cases. Am J Trop Med , 21: 35–48, 1941.
29 FAUST, C . Some modern conceptions of amebiasis Science, 99: 45–51, 69–72, 1944
30 FAUST, E C , SAWITZ, W , TOBIE, J , ODOM, V AND LINCICOME, D. R.: Comparative efficiency of various technics for the diagnosis of protozoa and helminths in the feces J. Parasitol., 25: 241–262, 1939.
31 GOLDMAN, M Polyvinyl-alcohol-fixative method for shipping fecal smears. *Pub Health Lab* , 6: 38–39, 1948
32 HAMILTON, H. E , CARNEY, R. G AND ZAVALA, D. C Unpublished data.
33 HAMILTON, H E. AND ZAVALA, D. C Amebiasis in Iowa· diagnosis and treatment. J Iowa M Soc , 42: 1–6, 1952
34. HARDY, A V Dysentery in Korea Annual Meeting American Society of Tropical Medicine, Chicago, Illinois, Nov. 15–17, 1951
35 HASKINS, W T , LUTTERMOSER, G W AND BRADY, F. J : The physiological disposition of diodoquin, vioform, and chiniofon in the rabbit as determined with radioiodine Am J Trop Med , 30: 599–612, 1950
36 HOEKENGA, M T . A comparison of aureomycin and carbarsone in the treatment of intestinal amebiasis. Am. J. Trop Med , 31: 423–425, 1951

patient's complaints. Even in the absence of positive laboratory tests the patient should probably be retreated. Patients showing other signs of the disease should be retreated without delay. Post treatment stool examinations are recommended. The practical point arises as to when and how often should they be repeated. There is no absolute answer for this question. Depending upon the circumstances a complete single stool examination in conjunction with an evaluation of the patient's clinical state should be carried out at 2 and 6 month intervals. If these examinations are negative the patient can be considered clinically free of the disease. If there are findings suggestive of recurrence of the infection sigmoidoscopic examination and further studies must be carried out.

In our experience the complement fixation test may again become positive with recurrence of hepatitis. Additional treatment is indicated. If the patient recovers from a hepatitis and remains symptom-free yet the complement fixation test is still positive retreatment should be carried out. We have had a patient with a persistently positive complement fixation test for a year who has had four courses of treatment. At no time after the initial treatment have there been symptoms or signs of disease. We believe such a patient needs no further treatment but should be examined periodically for evidence of active disease

REFERENCES

1. Anderson, H. H Current therapy of amebiasis Am. Pract , 4: 218–221, 1949
2. Armstrong, T G , Wilmot, A J and Elsdon-Drew, R Aureomycin and amebic dysentery Lancet, 1: 10–12, 1950
3 Banker, D. D Amoebic abscess of the liver Indian Physician, 6: 254–262, 1947. (See also Trop Dis Bull , 45: 338–340, 1948)
4 Berberian, D A , Dennis, E W and Pipkin, C A The effectiveness of bismuthoxy p-N-glycolylarsanilate (milibis) in the treatment of intestinal amebiasis Am J Trop Med , 30: 613–623, 1950
5 Bohts, I H Personal communication
6 Boyers, L M Human amebiasis, a review of 700 cases expressing 1961 complaints Calif & West Med , 39: 397–401, 1933
7. Bozicevich, J Ambeiasis Panel, Discussion Am J Trop Med , 30: 154–157, 1950
8 Brooke, M M , Goldman, M and Johnson, S A Polyvinyl alcohol fixative as a preservative and adhesive for protozoa in dysenteric stools and other liquid materials J Lab & Clin Med , 34: 1554–1560, 1949
9 Calero, C M Aureomycin in the treatment of intestinal amebiasis Am J Trop Med , 31: 566–568
10 [illegible] experimental study [illegible] 107, 1948
11 [illegible] Am J
12 [illegible] tic amoe-
13. [illegible]

58. TERRY, L. L. AND BOZICEVICH, J.: The importance of the complement fixation test in amebic hepatitis and liver abscess. South. M J., 41: 691-702, 1948.
59 TOBIE, J. E, MOST, H, REARDON, L V. AND BOZICEVICH, J.: Laboratory results of the efficacy of terramycin, aureomycin and bacitracin in the treatment of asymptomatic amebiasis Am. J. Trop. Med, 31: 414-419, 1951.
60 TOBIE, J E, REARDON, L V., BOZICEVICH, J, SHIH, B., MANTEL, N. AND THOMAS, E: The efficiency of the zinc sulfate technic in the detection of intestinal protozoa by successive stool examinations Am J Trop Med, 31: 552-560, 1951.
61. WRIGHT, W. H The public health status of amebiasis in the United States as revealed by available statistics Am. J. Trop. Med., 30: 123-133, 1950
62 YOUNG, V. M., FELSENFELD, O, SHLAES, W. H, YOSHIMURA, T. AND STEIGMANN, F.: A study of laboratory methods for diagnosing Endamoeba histolytica and their application to 5,048 persons from the Chicago area. Am J Dig Dis, 18: 126-130, 1951.
63. ZAVALA, D. C AND HAMILTON, H E. The recognition and treatment of hepatic amebiasis Ann Int. Med. 36: 110-125, 1948
64 ZINNEMAN, H H.: Ten cases of amoebiasis with arthritic complaints. Am J. Dig. Dis, 17: 342-344, 1950

37. HOOD, M , SODEMAN, W. A AND AKENHEAD, W R.: Comparison of effectiveness of proctoscopic and stool examination for detection of amebiasis. Annual Meeting American Society of Tropical Medicine, Chicago, Illinois, Nov. 15-17, 1951.
38. HUSSEY, K L. AND BROWN, H W · The complement fixation test for hepatic amebiasis. Am. J Trop Med., 30: 147–154, 1950.
38a. KLATSKIN, G. AND FRIEDMAN, H,: Emetine toxicity in man Ann. Int. Med, 28: 892–915, 1948
39. LAIRD, R. L., DRINNON, V. P. AND DAVIS, A. B.: Routine culture methods in diagnosing Endamoeba histolytica. J Nat. Malaria Soc, 8: 198-201, 1948
40. LOEBER M AND D'ANTONI, J S Some recent experiences with amebiasis in children New Orleans Med & Surg J 100: 276–278, 1947
41 MCVAY, L V, LAIRD, R L AND SPRUNT, D. H : Treatment of amebiasis with aureomycin South M. J., 43: 308, 1951
42 MOST, H, MILLER, J. W AND GROSSMAN, E. J Treatment of amebiasis with bacitracin and other antibiotics Am J. Trop. Med, 30: 491–497, 1950
43. MOST, H, MILLER, J. W, GROSSMAN, E. J AND CONAN, N : Treatment of amebiasis with bacitracin J A. M A, 143: 792–794, 1950
44. MOST, H AND VAN ASSENDELFT, F.: Laboratory and clinical observations on the effects of terramycin in the treatment of amebiasis Ann. N. Y. Acad Sci, 53: 427–428, 1950
45. MOST, H AND VAN ASSENDELFT, F Laboratory and clinical observations on the effect of terramycin in the treatment of amebiasis Am J Trop Med, 31: 284–285, 1951
46 ORBISON, J A, REEVES, N, LEEDHAM, C L AND BLUMBERG, J M Amebic brain abscess Medicine, 30: 247–282, 1951
47 PERRY, M W *Non dysenteric amebiasis general considerations of possible relationship to rheumatoid arthritis* M Ann District of Columbia, 16: 119–125, 1947
48 PHILLIPS, B P Cultivation of E histolytica with T cruzi. Science III, No. 2871, p. 8, 1950
49. PHILLIPS, B P Comparative effects of certain species of Trypanosomidae on the growth of Endamoeba histolytica in the absence of bacteria Am J Trop Med, 31: 290–294, 1951
50 PHILLIPS, B P AND REES, C W Growth of E histolytica with live and heat-treated T cruzi Am J Trop. Med, 30: 185–191, 1950
51 RADKE, R A Treatment of amebiasis with atabrine combined with carbarsone Ann Int Med, 34: 1432–1444, 1951
52 RADKE, R A Amebiasis, evaluation of new therapies U S Armed Forces M J, 2: 1231–1234, 1951
53 [illegible] to intestinal [illegible]
54 [illegible] therapeutic [illegible] Int Med, 88: 752–759, 1951
55. SHUTE, D · Liver damage in amoebiasis Brit M J, 1: 172–175, 1947
56. SODEMAN, W A. Clinical picture of hepatic amebiasis Am J Trop Med, 30: 141–146, 1950
57. SODEMAN, W A, DOERNER, A A, GORDON, E M AND GILLIKIN, C M Chloroquine in hepatic amebiasis Ann Int Med, 35: 331–341, 1951

PRESENT CONCEPTS OF THE LIFE CYCLE OF THE MALARIA PARASITE

Before proceeding to detailed discussions of specific antimalarial drugs, it is desirable to review briefly present day concepts of the life cycles of those species of *Plasmodium* which produce human malaria The importance of understanding these concepts is obvious when it is noted that they not only played a major role in the development of the newer and more effective drugs, but currently guide the intelligent application of these compounds at the bedside and in the field.

Prior to 1944, the generally accepted conceptions of the life cycles of human plasmodia followed the often described pattern in figure 1 and were based on the classical discoveries of Laveran, Ross, and Grassi (31, 41, 46) and the purported findings of Schaudinn (47). In brief, these conceptions embraced an asexual or schizogonous cycle taking place in the erythrocytes of the human host and a sexual or sporogonous cycle initiated in human erythrocytes but carried to completion in the anopheline vector These cycles were linked by the process of mosquito biting, which on the one hand led to introduction of the sexual forms of the plasmodium into the mosquito gut with subsequent fertilization and development to the sporozoite stage, and on the other to introduction of mature sporozoites into the circulation of the human host and, as shown in the illustration, direct invasion of the erythrocytes by these forms.

The correctness of the major features of these cycles within vertebrate and invertebrate hosts has never been questioned seriously. As early as 1924, however, there were growing doubts about the concept of direct invasion of erythrocytes by sporozoites. Not only had competent investigators failed to confirm the original observations of Schaudinn, but a growing body of indirect evidence suggested that direct entry did not occur. Thus it was noted repeatedly that no matter how many sporozoites were contained in an infecting dose, a certain minimum number of days, varying somewhat with the different species of human plasmodia, elapsed before parasites could be detected in blood by microscopic examination or before clinical evidence of malaria could be obtained. In addition, it was observed that for some days after sporozoite inoculation, blood taken from the inoculated subject did not give rise to infection when injected into susceptible recipients These observations were in striking contrast to those obtained in transfusion malaria where infections were produced by inoculation of erythrocytic parasites (trophozoites). Here the incubation period was always inversely related to the size of the parasite inoculum, and blood taken from the inoculated subject was at all times infectious when injected into susceptible recipients Finally it was found that doses of quinine which controlled or eradicated infections induced by inoculation of erythrocytic

The Present Status of the Chemotherapy of Human Malaria

L. H SCHMIDT, PH.D.

Although no longer regarded as a significant medical problem within continental United States, malaria still remains the most prevalent human disease, affecting, according to conservative estimates, no fewer than 700,000,000 of the world's inhabitants and exacting staggering tolls in terms of morbidity and mortality Whereas in many areas these tolls have been reduced or eliminated by measures designed to control or eradicate the anopheline vectors of the disease, such procedures are often difficult to apply effectively because of economic, geographic, or bionomic limitations. Unfortunately one or more of these obstacles appears to exist in those areas where the ravages of malaria are greatest. For the regular inhabitants of such regions and for those who for one reason or another must reside in these places for longer or shorter periods, chemotherapeutic measures aimed at protection of the human host offer the most practical hope of escaping the consequences of malaria infection

In view of the above situation, it is especially fortunate that there have been remarkable improvements in malaria therapeutics in recent years Prior to 1941, and for that matter for over 300 years, administration of cinchona products and specifically quinine was almost synonymous with malaria therapy Although this alkaloid possessed marked deficiencies in inherent activity and pharmacological characteristics, and was not available in sufficient quantities to meet global requirements, it nevertheless occupied an almost impregnable position until the circumstances of World War II abruptly eliminated the established sources of supply. These circumstances, coupled with the necessity of carrying on military operations in areas where malaria was hyperendemic, brought to the fore the search for a "quinine substitute" Stimulated immeasurably by a new understanding of certain of the basic biological characteristics of malaria, this search not only resulted in the finding of a suitable substitute but has led and is still leading to the development of a variety of synthetic drugs with therapeutic qualities far superior to those of quinine In the past five years, the most outstanding groups of these drugs have undergone critical evaluation, both in the laboratory and the field. It is the purpose of the present report to summarize the backgrounds, merits, and limitations of those drugs which have been subjected to the most intensive study and which appear to offer greatest promise.

parasites did not prevent infection when administered after sporozoite inoculation. This latter observation in particular led James (38) to discard the older concept of direct entry of sporozoite into erythrocyte and to suggest that there was a tissue stage intermediate between the sporozoite and erythrocytic parasite

This theory soon received confirmation from work in the avian malarias; first indirectly, through demonstration via subinoculation procedures of the existence of infective material in organs during the days immediately following introduction of sporozoites, at the time when the blood was non-infectious (62); and finally, directly, through essentially a complete description of the sequence of changes which occurs in the transformation of the sporozoites of *P. gallinaceum* to erythrocytic parasites (36). This latter work as well as numerous collateral studies in a variety of avian malarias showed conclusively that sporozoites left the blood soon after inoculation, entered cells of the lymphoid-macrophage system and there underwent a series of developmental changes prior to invasion of the bird erythrocytes

This direct demonstration in the avian malarias of the existence of tissue forms intermediate between sporozoite and erythrocytic parasite was soon followed by similar demonstrations in primate malaria, first in *P. cynomolgi* infections in the rhesus monkey (15, 34, 57), then in *P. vivax* and *P. falciparum* infections in man (55, 57) With all three plasmodia, forms believed to be intermediate between sporozoite and erythrocytic parasite have been found in the livers of the vertebrate hosts.

The finding of pre-erythrocytic forms as described above filled a long-standing void in knowledge of the early development of the malaria parasite. It did not, however, provide an explanation for the repeated relapses which are characteristic of sporozoite-induced infections with *P vivax* or *P malariae* exposed to presumably adequate chemotherapeutic treatment. Borrowing again from certain analogies in the avian malarias, these relapses have been attributed to the activity of persisting pre-erythrocytic stages or of exoerythrocytic* tissue stages derived therefrom Although supported by an overwhelming body of indirect evidence similar to that which pointed to the presence of pre-erythrocytic tissue stages, this concept has not yet been proved directly in the human malarias. Such proof has been obtained, however, in *P cynomolgi* infections in the rhesus monkey, a disease which for practical purposes is the simian counterpart of *P. vivax* infections in man Two groups of investigators (16, 56) have found forms similar to pre-erythrocytic stages in the livers of rhesus monkeys with long-standing infections with the above plasmodium. It seems reasonable to believe that

* The term exoerythrocytic refers to tissues other than blood

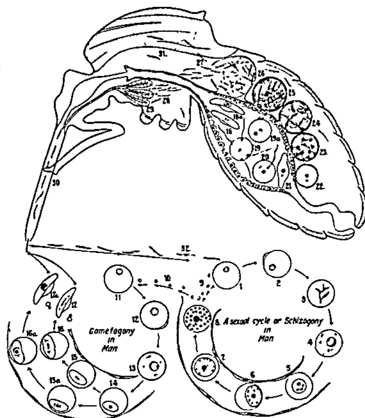


FIG 1 CONCEPTIONS OF SPOROGONOUS AND SCHIZOGONOUS CYCLES OF HUMAN PLASMODIA DERIVED FROM ORIGINAL OBSERVATIONS OF LAVERAN, ROSS, SCHAUDINN, AND OTHERS

1 to 8 Development of asexual parasites in the erythrocytes of the mammalian mature merozoites

to plasma, destined to invade normal

nto plasma, destined to develop into sexual parasites

gametes illustrated)

18 and 18a Mature male and female gametes ingested by mosquito during biting process

parasites did not prevent infection when administered after sporozoite inoculation This latter observation in particular led James (38) to discard the older concept of direct entry of sporozoite into erythrocyte and to suggest that there was a tissue stage intermediate between the sporozoite and erythrocytic parasite.

This theory soon received confirmation from work in the avian malarias; first indirectly, through demonstration via subinoculation procedures of the existence of infective material in organs during the days immediately following introduction of sporozoites, at the time when the blood was non-infectious (62), and finally, directly, through essentially a complete description of the sequence of changes which occurs in the transformation of the sporozoites of *P. gallinaceum* to erythrocytic parasites (36). This latter work as well as numerous collateral studies in a variety of avian malarias showed conclusively that sporozoites left the blood soon after inoculation, entered cells of the lymphoid-macrophage system and there underwent a series of developmental changes prior to invasion of the bird erythrocytes

This direct demonstration in the avian malarias of the existence of tissue forms intermediate between sporozoite and erythrocytic parasite was soon followed by similar demonstrations in primate malaria, first in *P. cynomolgi* infections in the rhesus monkey (15, 34, 57), then in *P vivax* and *P. falciparum* infections in man (55, 57) With all three plasmodia, forms believed to be intermediate between sporozoite and erythrocytic parasite have been found in the livers of the vertebrate hosts.

The finding of pre-erythrocytic forms as described above filled a long-standing void in knowledge of the early development of the malaria parasite It did not, however, provide an explanation for the repeated relapses which are characteristic of sporozoite-induced infections with *P. vivax* or *P. malariae* exposed to presumably adequate chemotherapeutic treatment Borrowing again from certain analogies in the avian malarias, these relapses have been attributed to the activity of persisting pre-erythrocytic stages or of exoerythrocytic* tissue stages derived therefrom. Although supported by an overwhelming body of indirect evidence similar to that which pointed to the presence of pre-erythrocytic tissue stages, this concept has not yet been proved directly in the human malarias. Such proof has been obtained, however, in *P cynomolgi* infections in the rhesus monkey, a disease which for practical purposes is the simian counterpart of *P. vivax* infections in man Two groups of investigators (16, 56) have found forms similar to pre-erythrocytic stages in the livers of rhesus monkeys with long-standing infections with the above plasmodium. It seems reasonable to believe that

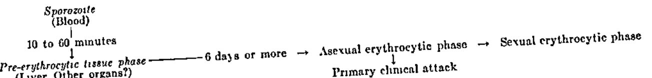
* The term exoerythrocytic refers to tissues other than blood

when inherent technical difficulties are surmounted, such forms will also be demonstrated in infections with *P. vivax* and *P. malariae*.

The above findings make it possible to describe the human malarias in terms of the developmental phases of the invading plasmodia. Such a description, as it applies to falciparum and vivax malaria, is set forth in figure 2. This figure indicates that in falciparum malaria there are essentially two different infections. The first of these involves tissues other than blood, produces no apparent adverse reactions in the host, but leads directly to a second infection which involves the erythrocytes and produces all of the symptoms and adverse reactions associated with clinical malaria These same two infections are common to the early phases of vivax malaria. In this disease, however, there is an additional infection of tissues other than blood, which arises from the original tissue infection at or about the time blood infection is established This late tissue infection, like the earlier pre-erythrocytic infection, is also devoid of adverse effects on the host but is capable of reactivating erythrocytic infection to produce clinical symptoms and what is termed relapse. The developmental phases and disease mechanisms of infections with *P. malariae*, although not indicated in figure 2, doubtless follow essentially the pattern assigned to vivax malaria.

Although the present review is not concerned primarily with problems of the life cycle of the malaria parasite, it would be unwise to leave this discussion without pointing to certain deficiencies in the concepts outlined above. For example, there is still no unanimity of opinion as to the actual locus of pre-erythrocytic or late exoerythrocytic development. It is an open question whether such development occurs exclusively in the liver, and, if so, only in the parenchymal cells, or whether it occurs throughout the body in cells of the lymphoid-macrophage system. Likewise, there is no indication whether the development of sporozoite to erythrocytic parasite is a single stage process or whether such development is a sequence of distinctly different processes similar to those encountered in certain avian malarias. The question of the nature of the exoerythrocytic forms responsible for relapse poses another problem, for it has not yet been determined whether such forms are merely persisting pre-erythrocytic stages or whether they are the progeny of a separately maintained exoerythrocytic cycle The frequency of maturation of the exoerythrocytic stages into forms capable of invading the erythrocytes and the factors controlling this matu- [illegible] broad varia- [illegible] t strains [illegible] e issues, and others seemingly less important at the moment, may well modify our present concepts of therapy just as did the original recognition of the existence of an exoerythrocytic cycle

2. SCHEMATIC REPRESENTATION OF DEVELOPMENTAL PHASES OF *P. falciparum* AND *P. vivax* MALARIAS IN HUMAN HOST



TYPES OF ANTIMALARIAL ACTIVITY REQUIRED FOR CONTROL OF THE VARIOUS HUMAN MALARIAS

The conceptions of the life cycle of the malaria parasite outlined above now make it possible to define approaches to chemotherapy of the disease as relatively discrete processes. According to these conceptions, falciparum malaria may be controlled by drugs which are either *prophylactic* or *schizonticidal*. In the first instance the attack is against the pre-erythrocytic stages of the parasite, in the second against the asexual erythrocytic stages, either before or after clinical disease occurs In either case, successful attack on the erythrocytic parasites is synonymous with eradication of the disease. The situation is somewhat more complicated in the case of vivax malaria, and probably so in infections with *P. malariae*. Attack on the pre-erythrocytic phases obviously controls the infection; attacks on the erythrocytic phases cause only temporary remission of clinical disease. Once pre-erythrocytic development is completed, total control of vivax malaria can be achieved only by measures aimed at both eradication of the erythrocytic parasites (*schizonticidal*) and elimination of the underlying tissue stages (*curative*)

Thus complete chemotherapeutic control of malaria requires drugs which possess activities against pre-erythrocytic, erythrocytic, and late exoerythrocytic phases of the human plasmodia. To date no one drug has been found which meets these requirements completely As noted below, however, a close approach to this ideal was achieved in one drug (chlorguanide) and practical achievement accomplished by a combination of one of several schizonticidal drugs with the most effective of the 8-aminoquinolines.

THE ATTRIBUTES OF CURRENTLY AVAILABLE ANTIMALARIAL DRUGS

There are probably few diseases where therapeutic evaluations in a natural habitat present as many problems as malaria Human malaria is commonly regarded as three separate diseases caused by three different species of *Plasmodium*, each with special biological characteristics, only a few of which have been described in this report It must also be recognized that each major species is made up of many different strains, which in [illegible], virulence, and [illegible] almost as broad [illegible] posed by these factors are complicated even further by coexistence in the same subject of infections with two or more plasmodial species, gross variations in both acquired and natural immunity of the host, and uncertainty as to the weight or duration of infection. Add to these the difficulties of achieving regular drug administration, of follow-up required for determining the

degree of therapeutic success, and complications imposed by intercurrent disease and one may readily understand why it is so difficult to obtain definitive evaluations of antimalarial drugs through field studies in areas where malaria is most prevalent, despite an almost unlimited supply of clinical material.

It is appropriate to note that the rapid development of malaria therapy in recent years has been stimulated and implemented by three fortuitous circumstances: 1) the discovery of the value of malaria fever in the treatment of general paresis; 2) the availability of large numbers of human volunteers as a consequence of the fervor of World War II; and 3) the movement, under fairly well controlled conditions, of large numbers of non-immune individuals into and out of areas where malaria is endemic. These events have made it possible to appraise the properties of potentially useful drugs in a controlled manner with due regard for many of the factors noted in the preceding paragraph The consequences of this approach have been threefold: 1) to provide definitive assessments of the properties of new drugs in a way which can be achieved only in the laboratory (21, 53), 2) to eliminate rapidly large numbers of drugs of second rate value, and thus concentrate efforts on relatively few compounds; 3) to bring drugs to trial on native populations with basic properties defined and only general utility remaining to be determined. The present review leans heavily on these controlled investigations

Only five drugs or groups of drugs, of the 100-odd candidate compounds of the past ten years, have achieved a position of sufficient importance to merit discussion here These drugs are: 1) quinine and other cinchona alkaloids, 2) quinacrine, 3) chloroquine and two related 4-aminoquinolines, 4) chlorguanide, and 5) a group of 8-aminoquinolines, of which primaquine is the most useful The chemical structures of these compounds together with some of the synonyms or trade names under which they appear in the literature have been set forth in figure 3. A detailed description of dosages of these drugs now recommended for diverse uses will not be attempted here. Such information can be obtained from numerous reports including the excellent synopsis by Cooper (13).

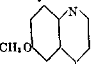
Quinine and Cinchona Alkaloids

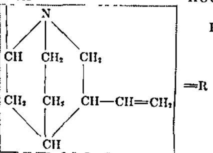
Although quinine and mixtures of crude cinchona alkaloids now occupy a position of decreasing importance in malaria therapeutics and in the foreseeable future may cease to play any role, their inclusion in the present discussion seems indicated if for no other reason than that use over several hundred years has provided standards of accomplishment against which other drugs may be measured In addition there are still areas where ad-

FIG. 3. CHEMICAL STRUCTURES OF AVAILABLE ANTIMALARIAL DRUGS

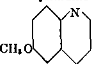
(1) Quinine and other cinchona alkaloids

Quinine

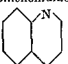




Quinidine



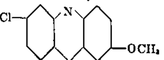
Cinchonidine

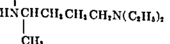


Cinchonine



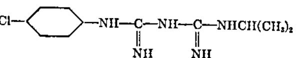
(2) Quinacrine





Synonyms Mepacrine (B P.)
Atabrine
Atebrin
Acriquine
Erion

(4) Chlorguanide



Synonyms: Proguanil (B P)
Paludrine
Palusil
Guanatol

(3)
4 aminoquinolines

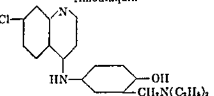
Chloroquine

Cl, N

$HN\,CH(CH_3)CH_2CH_2CH_2N(C_2H_5)_2$

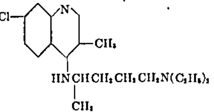
Synonyms SN 7618
Resochin
Aralen
Nivaquine B

Amodiaquin



Synonyms SN 10,751
Camoquin
CAM-AQ1

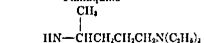
Sontoquine



Synonyms. SN 6911
Sontoquin
Santoquine
Nivaquine
Nivaquine C

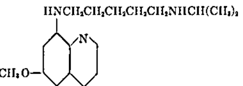
(5)
8-aminoquinolines

Pamaquine



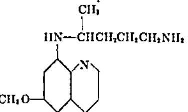
Synonyms: Plasmochin
Plasmoquine
Praequine

Pentaquine



Synonym: SN 13,276

Primaquine



Synonym. SN 13,272

ministration of quinine or totaquine is routine practice, either because of economic considerations which restrict purchase of more effective but more expensive drugs, or because of therapeutic conservatism.

The sequence of events leading to the use of quinine in malaria requires no comment. Analysis of the antimalarial characteristics of this alkaloid shows that it possesses schizonticidal activity but has no effect on development of pre-erythrocytic parasites of any malaria species or on the activity or survival of the late exoerythrocytic forms of vivax malaria. Thus, in a strict sense quinine is neither prophylactic nor curative. However, as years of use testify, when properly administered the drug is a comparatively effective schizonticide. Given repeatedly to individuals who are continuously exposed to the bites of infected mosquitoes, quinine will in most instances keep parasitemia below levels where clinical evidence of malaria is apparent and in many instances will destroy erythrocytic parasites as quickly as they appear. When clinical disease is already present, administration of substantially larger doses will in the majority of cases eradicate parasitemia and abolish symptoms of malaria Quinine attacks the erythrocytic parasites at all stages of development. Consequently, both parasitemia and fever are promptly reduced, complete clearance of erythrocytic parasites is considerably less rapid and is seldom achieved by less than 4 days of treatment in vivax malaria and 7 days of therapy in infections with *P. falciparum* Since parasite eradication is synonymous with cure in falciparum malaria, this disease may be cured by quinine assuming that reinfection does not take place Under similar conditions a high percentage of vivax infections (70 to 100) may be expected to relapse, in many cases within 7 to 10 days after termination of quinine therapy.

As mentioned previously, there are serious limitations to the usefulness of quinine. Probably most important are the wide variations in the susceptibility of different species and various strains of *Plasmodium* to the schizonticidal action of this compound These variations occur both in vivax and falciparum malaria but are particularly important in the latter where the consequence of inadequate treatment may be death One could cite innumerable examples of these variations which have occurred both in the routine suppression of infection and in the treatment of clinical attacks It is sufficient to refer to the results of the systematic studies of Shannon (52) and Shute (58) The former found that the total dose of quinine required to control infections with two strains of *P falciparum* varied by a factor of two while that required to control infections with two strains of *P. vivax* varied by a factor of four Shute reported a tenfold difference in the total dose of quinine required to control infections with Indian and Italian strains of *P falciparum*. These variations in strain response are

serious even when dealing with a population which has a fairly high degree of immunity. They can have disastrous consequences when the concern is protection of non-immunes.

The toxicity of quinine is a problem of much importance, particularly in view of the broad variations in strain susceptibility noted above. The drug possesses a relatively low therapeutic index (Toxic Dose/Effective Dose) thus making it practically impossible to expand dosages over wide limits Daily doses, equivalent to 2 grams of quinine base per day, frequently necessary to control active infections and in some cases subeffective, are close to the maximal tolerated level for most individuals and are productive of unpleasant toxic symptoms in others. The reactions to overdosage with quinine include not only convulsions, cardiac irregularities, nausea, and vomiting, but also effects on the optic and auditory nerves, the latter effects often irreversible. Although quinine blindness is not a common occurrence, deafness, particularly in children, is not an uncommon finding To these undesirable qualities must be added the local tissue toxicity of quinine which is a serious liability to the use of parenterally administered quinine in cerebral malaria where oral medication cannot be accepted. Intramuscular administration frequently produces local necrosis and sterile abscesses and for years has been generally condemned Intravenous administration of the drug by all but the most skilled often produces sloughs. A profound lowering of the blood pressure is another untoward reaction.

Whether a result of trial and error or an arbitrary practice, it has been the custom for years to administer quinine in divided daily doses. Recent studies have shown that this practice is imperative Thus it has been found that the antimalarial activity of quinine is a direct function of the plasma concentration, that closely spaced doses are required to maintain effective plasma concentrations (52), and that the drug is three times as effective when administered in a six hourly dosage schedule as when given twenty-four hourly, total dose of drug remaining the same (Canfield and Schmidt, unpublished observations) In addition the latter observation shows that effective therapy on a single daily dose regimen would be toxicologically impossible This need for frequent quinine administration is a serious burden when treatment of large population groups is involved, and is probably one of the drug's greatest disadvantages.

The apparent relation of quinine therapy to the massive intravascular hemolysis and hemoglobinuria termed blackwater fever is another factor which limits the usefulness of this alkaloid. At present it is not certain whether quinine per se is responsible for this syndrome or whether the hemolysis results from a series of cellular immune reactions associated with chronic but partially suppressed falciparum malaria. Whatever the

mechanism, general use of quinine is implicated, for substitution of the newer and more effective drugs has caused a phenomenal lowering of the incidence of blackwater fever if not elimination of this once serious problem.

Interest in the other cinchona alkaloids, quinidine, cinchonine, and cinchonidine, rests largely in the fact that they are major components of a low cost mixture called totaquine. These compounds have been investigated extensively by a number of workers (28, 60), who have reached the conclusion that dose for dose quinidine is several times more active than quinine, cinchonidine is about equally active whereas cinchonine is only one half as active The mixture of these alkaloids in totaquine has an antimalarial effect not materially different from that of quinine. In general the same factors which limit the usefulness of quinine restrict the application of the other cinchona alkaloids. The toxicity of cinchonidine is distinctly greater than that of quinine, particularly with respect to producing nausea and vomiting. Low cost would appear to be the only justification for the planned use of totaquine.

Quinacrine

Introduced into malaria therapeutics in 1930-31, quinacrine was the first drug which exhibited antimalarial properties superior to those of quinine This superiority was recognized in a few quarters by the mid-1930's, but was not generally appreciated until the circumstances of World War II precipitated general use of the drug Several factors were responsible for this delayed recognition of the usefulness of quinacrine Failure to understand the pharmacological properties of the compound was certainly important, as was the general willingness of malariologists to accept from any new drug a therapeutic accomplishment comparable to that of quinine rather than to exploit such a substance to its ultimate limits

An understanding of certain of the pharmacological characteristics of quinacrine is so important to the use of this drug, that these properties should be considered at this point. Insofar as distribution in the body and metabolism are concerned, the behavior of quinacrine is wholly unlike that of quinine. The latter compound is rapidly absorbed from the gastrointestinal tract, localized to only a minor extent in any organ or tissue, extensively degraded and quickly excreted Quinacrine is also rapidly absorbed, but is metabolized and excreted extremely slowly, 10 per cent or less of a given dose appearing in the urine each day. It is localized extensively in all organs and tissues, but particularly in the liver, spleen, lung, skin, and intestinal mucosa, where it attains concentrations at least several hundred times those in plasma. Tissue affinity is so great that three to four weeks are required for complete clearance of the compound from the body after administration of quinacrine is terminated Failure to appreciate this tissue

affinity and to devise practical means of overcoming it was responsible for the pre-1943 impression that quinacrine was slower acting than quinine. In 1943 it was recognized (54) that the antimalarial activity of quinacrine was closely related to the amount of the drug in plasma and that concentrations of the drug in blood of a magnitude necessary for rapid therapeutic effects could be achieved by the simple expedient of administering larger priming doses

With respect to specific antimalarial properties, quinacrine, like quinine, is strictly a schizonticide As shown by the classical studies of Fairley (26), the drug has no direct effects on the pre-erythrocytic development of either *P falciparum* or *P. vivax* or on the late exoerythrocytic development of the latter plasmodium. When its administration in laboratory-type studies is limited to the incubation period, quinacrine causes a delay in the appearance of parasitemia and clinical symptoms in vivax malaria and either delay or disease prevention in falciparum malaria. This finding, first interpreted as an effect of the drug on pre-erythrocytic development, is now recognized as due to retention of quinacrine in tissue in sufficient amounts to maintain schizonticidal concentrations in the blood for extended periods A similar explanation accounts for the lengthened remission periods that characterize established vivax infections which have been treated with quinacrine. At one time, however, this effect was regarded as evidence of action of the drug on the forms of *P vivax* responsible for relapse

In actual practice, administration of quinacrine in repeated daily doses of 0 1 gram of the dihydrochloride, or in twice weekly doses of 0 4 to 0.5 gram, to individuals continuously exposed to the bites of infected mosquitoes, has been remarkably effective in preventing the appearance of parasitemia and clinical symptoms With the exception noted below, these regimens usually effect complete protection against falciparum malaria as evidenced by the continued absence of infection when ingestion of quinacrine is terminated Regular protection of this order is not achieved, however, in the case of vivax infections which become active weeks to months after withdrawal of the drug, the precise time of activation being a function of the infecting strain. Since quinacrine is effective against all stages of the developing trophozoites, the administration of this drug in active infections with either *P. vivax* or *P. falciparum* causes a prompt reduction of parasitemia and clinical symptoms, and effects cure in the case of falciparum malaria when opportunities for reinfection are precluded. Such results can usually be achieved by administering a total dose of 2 8 grams of the dihydrochloride salt of quinacrine over a 7-day period, with 1 0 gram given in divided doses on the first day and 0 3 gram in divided doses on each of 6 succeeding days

The results of numerous studies indicate that there are significant varia-

tions in the effectiveness of quinacrine against infections with different strains of malaria parasites. Unlike the situation with quinine, however, the schizonticidal activity of quinacrine is sufficiently great so that when used as described this drug usually effects satisfactory suppression of parasitemia or control over the clinical attack. One important exception to this must be noted During the World War II campaigns, Fairley (24) encountered infections with a strain of *P falciparum* in a very localized area of New Guinea, which could not be suppressed by the daily administration of 0.1 gram of quinacrine hydrochloride, and only partially suppressed by daily doses of 0.2 gram. This must be emphasized as a phenomenon of natural rather than acquired resistance.

It should be noted that intramuscular administration of quinacrine provides rapid and effective control of cerebral malaria Administered via this route, the drug produces some local tissue reaction but far less than that produced by quinine. Intravenous administration of quinacrine may lead to serious convulsive seizures and should not be employed unless the drug is highly diluted in saline or other parenteral fluid.

It is evident from this brief summary that quinacrine is superior to quinine in a number of important respects. Inherently greater antimalarial activity, resulting in more reliable suppressive action and more certain and prompt control of the clinical attack, is an important point; less frequent drug administration is another. To these should be added the more certain control of cerebral malaria, and a pronounced reduction in the incidence of blackwater fever Despite these advantages it would be erroneous to infer that quinacrine is an ideal drug even as a schizonticide It must still be administered in daily doses or at best twice weekly for suppressive purposes, and for periods of 7 days for complete control of clinical attacks. It dyes the skin yellow, which although only of cosmetic importance, limits acceptability of the drug by some persons In addition quinacrine exhibits certain toxic properties even at dosages in general use. Initial use not infrequently produces symptoms of gastrointestinal distress comprising nausea, vomiting, abdominal cramps, and diarrhea. Fortunately, these symptoms disappear with continued drug administration. Symptoms of cerebral irritation are not uncommon, *these may vary in degree from depression and anxiety* to restlessness and excitement bordering on hysteria. These symptoms disappear on withdrawal of the drug In humid tropical areas the occurrence [illegible] results [illegible]

ant reaction. It must be emphasized, however, that contrary to [illegible] which prevailed prior to World War II, symptoms of quinacrine intoxication, during ordinary usage, are less *frequent and less annoying* than those of quinine

Chloroquine and Other 4-aminoquinolines

The introduction of the 4-aminoquinolines and particularly chloroquine probably constitutes the most significant practical achievement in malaria therapeutics of the past ten years. In view of present recognition of the attributes of these drugs, it seems remarkable that they evoked so little early interest even among groups responsible for their preparation. The earliest work on the 4-aminoquinolines was an outgrowth of attempts to relate the antimalarial activities of quinine and quinacrine to certain structural features This effort resulted in the synthesis of two series of 4-aminoquinolines, one of which contained a chloro substituent at position 7 of the quinoline ring, the other a methoxy substituent at position 6. Chloroquine, one of the 7-chloro derivatives, was synthesized in Germany in 1934, but was discarded as too toxic after superficial examination in man. The first 4-aminoquinoline to receive a reasonably careful study was sontoquine, the 3-methyl derivative of chloroquine This compound was given limited clinical trial in Germany and was examined more extensively by the French, who used the drug in North Africa during World War II and found it to have high antimalarial activity and to be well-tolerated. These observations, coming to light following recovery of North Africa from the Axis Powers, stimulated interest in the 4-aminoquinolines in the United States The result was independent synthesis and examination in lower animals of a large series of derivatives, nine of which possessed features of sufficient interest to justify limited but systematic study in man. From this work (5), four compounds emerged as superior to the others. These were chloroquine, and amodiaquin (cf fig 3), oxychloroquine (7-chloro-4-3-diethylamino-2-hydroxypropylamino)-quinoline), and SN 9584 (7-chloro-4-(3-diethylaminopropylamino)-quinoline). Further small scale clinical studies indicated that chloroquine was somewhat superior to other drugs in this group, both in antimalarial activity and pharmacological properties. Consequently, it was selected for field trials and large scale production. There is now a large body of information on the use of chloroquine in various parts of the world, much of which deals with highly controlled studies

The pharmacological properties of chloroquine are grossly similar to those of quinacrine The drug is localized extensively in the tissues, and is metabolized and excreted slowly. Its localization in various tissues closely resembles that of quinacrine except that less chloroquine is found in liver and more in brain It is metabolized somewhat more slowly than quinacrine to yield degradation products, at least one of which has antimalarial activity similar to that of the parent drug On equivalent doses the concentrations of chloroquine in plasma are higher and better sustained than those of quinacrine This property of persistence coupled with phenomenal schiz-

onticidal activity makes it possible to achieve routine suppression of malaria by administration of chloroquine in single weekly doses, and to effect satisfactory control of active infections with from one to three days of drug administration. In the latter instance, prompt control of infection is facilitated by use of priming doses

The effectiveness of chloroquine as an antimalarial rests largely on its schizonticidal properties which are probably greater and more reliable than those of any known antimalarial drug Like quinacrine and quinine, chloroquine does not affect *pre-erythrocytic development of any of the human* plasmodia and does not prevent establishment of the late exoerythrocytic infection of *P. vivax*. It is sufficiently effective as a schizonticide to suppress infections completely when administered in single weekly doses of 0 3 gram base (0 5 gram of phosphate salt) Such intermittent dosage provides *complete protection against falciparum infections* but not vivax, which become active some weeks after chloroquine suppression is terminated In developed malaria, single doses of 0.6 gram base are sufficient to eradicate many active infections with *P. falciparum* and to eliminate most clinical attacks of vivax malaria, particularly in persons with partial immunity. In essentially all cases, effective treatment of active infections may be achieved with a dosage of 1.5 grams of base (2.5 grams of phosphate salt), 0 6 gram administered initially, 0 3 gram 6 hours later and on each of two succeeding days. This dosage eradicates falciparum infections in all cases but does not prevent relapses due to *P. vivax* Such relapses generally occur 5 to 8 weeks or even longer after treatment of the acute attack Thus remission periods are strikingly longer after chloroquine treatment than after use of quinacrine These protracted remission periods are probably due to tissue localization of the drug which in turn maintains schizonticidal levels of chloroquine in the circulating blood.

Like quinine and quinacrine, chloroquine attacks the erythrocytic parasites at all stages of development This property together with high schizonticidal activity makes chloroquine the most rapidly acting of all known antimalarial drugs Its use in active infections usually results in elimination of fever in 24 hours and eradication of parasitemia in 48 to 72 hours. Because of this rapid action, chloroquine is especially useful in cerebral malaria. When administered intramuscularly it produces significantly less local tissue reaction than quinacrine Rapid and reliable absorption from the muscles makes intravenous medication unnecessary Thus far occurrence of blackwater fever during chloroquine administration has not been reported.

There are undoubtedly variations in the susceptibility of different strains of plasmodia to chloroquine Such differences have not been brought to light as yet, probably because the drug has always been employed in doses

well above the minimal effective level. Chloroquine has been used suppressively and therapeutically in essentially every part of the world where malaria occurs. To date there are no documented records of failure where doses such as the above have been employed.

Probably the greatest advantage of chloroquine is its high therapeutic index. The drug possesses extremely low toxicity at doses employed for suppression and therapy Weekly doses of 0 3 gram, which provide effective suppression, produce no significant reactions. As a matter of fact in one carefully controlled study (35) the incidence of reported reactions with placebos was greater than that with suppressive doses of chloroquine. In a group of 20 volunteers daily doses of 0 3 gram for ten consecutive weeks produced no noteworthy reactions (3). Full therapeutic doses of chloroquine in active infections have been reported to produce headache, pruritus, and blurring of the vision. These symptoms disappear promptly after the end of treatment Chloroquine is colorless and does not stain the skin.

As a result of early studies (18) on human volunteers, two of whom developed skin lesions not unlike the atypical lichen planus produced by quinacrine, considerable attention has been given to the possibility that chloroquine may induce a similar reaction No other occurrence of lesions of this type has been reported to date. Moreover, chloroquine has been administered uneventfully to many patients who had previously developed dermal lesions during suppressive use of quinacrine

On the basis of the above discussion it may be concluded that chloroquine is highly effective in the suppression and treatment of human malarias. Its only limitations appear to be cost and supply.

Amodiaquin (*Camoquine*), another 4-aminoquinoline, available in some parts of the world, is very similar to chloroquine in pharmacological and antimalarial characteristics. Whereas the most critical work (5) in human volunteers suggests that it is slightly inferior to chloroquine in activity and slightly more toxic, the differences are probably too small to have practical significance On the basis of present field experience, there is little to choose between the two drugs. It must be emphasized, however, that in contrast to experience with chloroquine, that with amodiaquin is relatively limited.

Sontoquine does not occupy as favorable a position as either of the above 4-aminoquinolines This drug is distinctly less active than chloroquine and probably not significantly more effective than quinacrine. Dose for dose it is slightly less toxic than chloroquine but this difference is vitiated by the need for larger doses of sontoquine to achieve the same suppressive or therapeutic effect. The compound is mentioned here only because it is marketed in certain areas and because a few workers have extolled its superiority over quinacrine

onticidal activity makes it possible to achieve routine suppression of malaria by administration of chloroquine in single weekly doses, and to effect satisfactory control of active infections with from one to three days of drug administration In the latter instance, prompt control of infection is facilitated by use of priming doses.

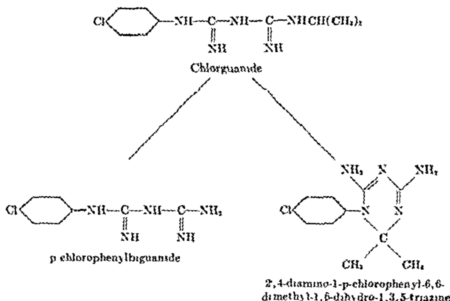
The effectiveness of chloroquine as an antimalarial rests largely on its schizonticidal properties which are probably greater and more reliable than those of any known antimalarial drug. Like quinacrine and quinine, chloroquine does not affect pre-erythrocytic development of any of the human plasmodia and does not prevent establishment of the late exoerythrocytic infection of *P. vivax*. It is sufficiently effective as a schizonticide to suppress infections completely when administered in single weekly doses of 0 3 (? gram base (0.5 gram of phosphate salt). Such intermittent dosage provides complete protection against falciparum infections but not vivax, which become active some weeks after chloroquine suppression is terminated. In developed malaria, single doses of 0 6 gram base are sufficient to eradicate many active infections with *P. falciparum* and to eliminate most clinical attacks of vivax malaria, particularly in persons with partial immunity. In essentially all cases, effective treatment of active infections may be achieved with a dosage of 1.5 grams of base (2.5 grams of phosphate salt), 0 6 gram administered initially, 0 3 gram 6 hours later and on each of two succeeding days. This dosage eradicates falciparum infections in all cases but does not prevent relapses due to *P. vivax* Such relapses generally occur 5 to 8 weeks or even longer after treatment of the acute attack Thus remission periods are strikingly longer after chloroquine treatment than after use of quinacrine. These protracted remission periods are probably due to tissue localization of the drug which in turn maintains schizonticidal levels of chloroquine in the circulating blood

Like quinine and quinacrine, chloroquine attacks the erythrocytic parasites at all stages of development This property together with high schizonticidal activity makes chloroquine the most rapidly acting of all known antimalarial drugs Its use in active infections usually results in elimination of fever in 24 hours and eradication of parasitemia in 48 to 72 hours. Because of this rapid action, chloroquine is especially useful in cerebral malaria. When administered intramuscularly it produces significantly less local tissue reaction than quinacrine Rapid and reliable absorption from the muscles makes intravenous medication unnecessary Thus far occurrence of blackwater fever during chloroquine administration has not been reported

There are undoubtedly variations in the susceptibility of different strains of plasmodia to chloroquine Such differences have not been brought to light as yet, probably because the drug has always been employed in doses

In considering the potentialities of chlorguanide in human malaria it is desirable to divide the discussion into two parts, one dealing with the early appraisal of the drug's potentialities, the other a reappraisal based on some 5 years of field performance. The first human use of chlorguanide (1, 22, 25, 26, 43) showed that this compound possessed a unique order of activity against the erythrocytic forms of *P. vivax*. Thus single daily doses as low as 10 mg, administered for 14 days in active infections effectively controlled clinical symptoms and eliminated parasitemia. Subsequent studies confirmed this finding and showed that a total dose as small

Fig 4 Metabolic Transformation of Chlorguanide



as 50 mg given in 4 days, or 100 mg in 10 days, would achieve the above effects. At the same time it was noted that the drug was highly effective in controlling active infections with *P. falciparum* although in some instances the total dose required to control this malaria was 5 to 20 times that needed in infections with *P. vivax*. This early work provided no information on the capacity of chlorguanide to eradicate established infections with either plasmodial species. Subsequent studies on active infections showed that the drug did eradicate falciparum malaria but did not prevent relapses in individuals infected with *P. vivax* even when the daily dose of drug was increased to 100 times the amount required to eliminate the erythrocytic phase of the infection.

Shortly after the initial demonstration of the schizonticidal action of

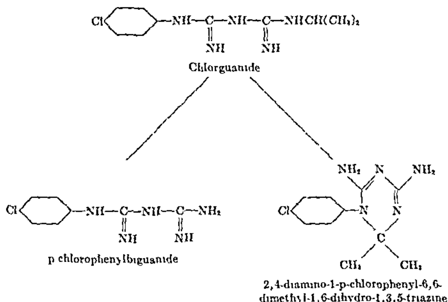
Chlorguanide

Chlorguanide (known almost exclusively outside of the United States by its trade name *Paludrine*), if not the most effective, is probably the most interesting and remarkable of all developments in modern malaria chemotherapy. The work which led to synthesis of this compound has been summarized completely and in a masterful manner by Curd, Davey, and Rose (20), and by Rose (45), and need not be related here It should be noted however, that chlorguanide was a direct outgrowth of interests in the antimalarial properties of the sulfanilamidopyrimidines, specifically sulfamethazine In structure (cf. fig. 3) this compound is a complete departure from quinoline derivatives considered thus far and from all other known antimalarial drugs.

Chlorguanide differs markedly from either quinine or quinacrine and the 4-aminoquinolines with respect to physiological disposition. Chlorguanide is absorbed from the gastrointestinal tract fairly completely, although not rapidly. It is localized in the tissues to a greater degree than the cinchona alkaloids, but far less than quinacrine and the 4-aminoquinolines. In general, the amounts of chlorguanide found in the plasma and tissues bear a constant relation to the dose of drug ingested (51). Elimination of the compound from the body, which is essentially complete within 3 days of termination of treatment, follows one of several pathways. From 25 to 40 per cent of the drug ingested is excreted unchanged either in urine or feces The drug in the stool represents material which is excreted via the bile or directly through the mucosa of the large intestine rather than drug which has escaped absorption. Elimination of metabolic products (fig. 4) accounts for the remaining 60 to 75 per cent of the chlorguanide administered (19, and Schmidt, Hughes, and Crounse, unpublished observations) One of these, p-chlorophenylbiguanide, results from removal of the isopropyl group from the terminal nitrogen of the parent drug. The other compound, 2,4-diamino-1-p-chlorophenyl-6,6-dimethyl-1,6-dihydro-1,3,5-triazine (8), is an entirely new type of compound, formed by condensation of the nitrogen adjacent to the phenyl group and the isopropyl group of chlorguanide Isolation and identification of these products, as well as determination of their antimalarial properties, has been a matter of great interest, for from almost the first use of chlorguanide there have been suggestions that the parent drug was not in itself active but that it was converted to an active degradation product (30, 32, 33) It is important to note, therefore, that p-chlorophenylbiguanide is devoid of antimalarial activity (Schmidt, Hughes, and Crounse, unpublished observations) and that, whereas the

In considering the potentialities of chlorguanide in human malaria it is desirable to divide the discussion into two parts, one dealing with the early appraisal of the drug's potentialities, the other a reappraisal based on some 5 years of field performance. The first human use of chlorguanide (1, 22, 25, 26, 43) showed that this compound possessed a unique order of activity against the erythrocytic forms of *P. vivax*. Thus single daily doses as low as 10 mg., administered for 14 days in active infections effectively controlled clinical symptoms and eliminated parasitemia. Subsequent studies confirmed this finding and showed that a total dose as small

FIG 4 METABOLIC TRANSFORMATION OF CHLORGUANIDE



as 50 mg given in 4 days, or 100 mg. in 10 days, would achieve the above effects At the same time it was noted that the drug was highly effective in controlling active infections with *P falciparum* although in some instances the total dose required to control this malaria was 5 to 20 times that needed in infections with *P vivax*. This early work provided no information on the capacity of chlorguanide to eradicate established infections with either plasmodial species Subsequent studies on active infections showed that the drug did eradicate falciparum malaria but did not prevent relapses in individuals infected with *P vivax* even when the daily dose of drug was increased to 100 times the amount required to eliminate the erythrocytic phase of the infection.

Shortly after the initial demonstration of the schizonticidal action of

chlorguanide, Fairley and coworkers reported the remarkable effects of the drug on pre-erythrocytic development. These investigators found that complete protection against infections with *P. falciparum* could be achieved with single doses of 50 to 100 mg. administered within 40 to 120 hours of the time of mosquito biting. However, when administered in this manner and even at doses of 1 gram per day for 10 days, the drug did not prevent establishment of *P. vivax* infections. It clearly had some effects on pre-erythrocytic development of this parasite, for in many cases the incubation period was prolonged for weeks after termination of chlorguanide administration, far beyond the expected period of drug retention It was also found that administration of chlorguanide to gametocyte carriers affected the sexual forms so that sporozoite development in the invertebrate host could not be completed.

To these remarkable antimalarial properties, the low toxicity of chlorguanide should be added. The drug is devoid of specific toxic effects on any organ system. When ingested in daily doses of 1 gram, moderate gastric distress and loss of appetite are the only significant reactions Doses larger than 1 gram produce severe nausea but no more serious reaction.

It is clearly evident, therefore, that the early assessments of the antimalarial properties and tolerability of chlorguanide provided ample grounds for enthusiasm. This drug had complete prophylactic action against one species of human plasmodium and exhibited partial prophylaxis against another, it possessed an order of schizonticidal activity greater than had been observed before; and it was well-tolerated. To these qualities were added low cost, no small consideration when long term administration to millions of persons is involved as in malaria The potentialities of chlorguanide as envisioned in 1946 are adequately summed up by the following quotation from Fairley: "Its pre-eminent therapeutic achievement is that in non-immunes a single dose of 100 mg. 1) may terminate a clinical attack of falciparum or vivax malaria, and 2) is a complete causal prophylactic* if given from 2 to 5 days after exposure to infective bites (*P falciparum*) As a complete causal prophylactic the dosage recommended is 100 mg every 3 or 4 days, i e, twice weekly (Wednesdays and Sundays). The potentialities of paludrine for the chemotherapeutic control of malaria in non-immune Europeans, as well as in indigenous native populations with premunity, are enormous, and call for controlled field investigations on a large scale."

As would be expected, these large scale field studies were not long in coming. Unfortunately their results have brought about a considerable deflation in early appraisals of the ultimate usefulness of chlorguanide.

* "Causal prophylactic" is synonomous with prophylactic and refers to destruction of pre-erythrocytic stages of the malaria parasite.

This deflation rests, first of all, on recognition that there are broad variations in the effectiveness of the drug against different strains of *P. falciparum* and *P. vivax*, and that there are particularly broad variations in the compound's activity against the erythrocytic forms of various strains of *P. falciparum*. Secondly, it has been recognized that the drug is relatively slow-acting against the erythrocytic parasites of all human plasmodia. And finally, it has been shown that the drug induces the development of resistant strains, the first demonstration of this phenomenon in the chemotherapy of human malaria

Variation in strain susceptibility to chlorguanide is an extremely perplexing problem showing no consistent pattern with respect to geographical distribution. In many areas of Africa, the drug has appeared to work well in suppressing malaria infections when given in twice weekly doses of 0.2 gram In other areas, daily doses of 0 2 gram are required for effective suppression. Similar variability in strain susceptibility has been found in southeast Asia and Indonesia. The variations in the ability of the drug to control active erythrocytic infections, particularly in falciparum malaria, are just as striking and far more serious. In some areas, single doses of 100 mg have controlled the active infection and apparently effected cure. In other areas, for example West Africa, daily doses of 900 mg for as long as 10 days have an uncertain therapeutic effect. In at least one instance (17) a strain of *P. falciparum* has been discovered in which pre-erythrocytic development can be controlled by extremely small doses of chlorguanide, while erythrocytic development is barely controlled with maximal tolerated doses.

The slowness of action of chlorguanide is a characteristic which was evident from almost the first use of the drug (39), but which was generally overlooked due to enthusiasm for other attributes of the compound. This slowness of action results from two now well defined properties of the drug: 1) an activity which is limited to one phase of erythrocytic development, and 2) a mode of action which is parasitostatic rather than parasiticidal. Thus, development of erythrocytic parasites is affected by chlorguanide only at the stage where the chromatin of the mature trophozoite undergoes cleavage to form the merozoites of the segmenter. Even this effect is a slow one. At first development of the parasite merely seems to be arrested at the above point; after a day or two these arrested parasites degenerate and are cleared from the blood through conventional phagocytic processes. The entire sequence of events from beginning of drug treatment to parasite clearance may require 5 to 8 days As might be anticipated, the speed of action of chlorguanide appears to be relatively independent of the amount of drug administered Thus where one is dealing with a susceptible strain, the therapeutic and parasitological response is as

rapid with a single dose of 100 mg. as it is with a dose of 1 gram. It must be re-emphasized that the mode of action of chlorguanide on erythrocytic parasites is distinctly *different from that of quinine, quinacrine, and the* 4-aminoquinolines. These latter compounds are parasitocidal and are effective against all phases of the erythrocytic parasite. The speed with which they produce therapeutic effects is related extremely closely to dosage. The slowness of action of chlorguanide limits the effectiveness of this drug in the treatment of cerebral malaria. It is not satisfactory for such purposes even when the infecting plasmodium is known to be susceptible to the drug. It should also be noted that the slowness of action of chlorguanide is so generally recognized that it has been officially recommended that the first day's treatment of an acute attack be supplemented by simultaneous administration of 900 mg. of quinacrine (12).

Development of chlorguanide resistance is a problem of far reaching implications The capacity of the drug to induce the development of resistant strains was first noted in the avian and simian malarias (6, 50, 64) In this work strains highly susceptible to chlorguanide were converted into wholly unsusceptible strains by successive treatment of persisting infections with gradually increasing doses of the drug. In the case of *P. cynomolgi* infections in the rhesus monkey a ten thousandfold change in the response of the parasite was effected within a period of three months. The resistant characteristic was a property of both asexual and sexual erythrocytic parasites and could be transmitted through the mosquito Chlorguanide-resistant strains of both *P falciparum* and *P vivax* have been prepared in the laboratory (14, 48, 49). As in the case of simian malaria, the resistant characteristic was transmitted through the mosquito Despite these find-
[illegible]

emerged in several areas where none had existed originally (9, 23, 29, 42, 61) The significant phase of this process is just beginning. In view of this, it is comforting to note that chlorguanide-resistant plasmodia are wholly susceptible to the 4-aminoquinolines and quinacrine (50)

In summary it may be said that although chlorguanide possesses remarkable properties, among which are ability to destroy the pre-erythrocytic forms of *P. falciparum* and to partially block development of such forms of *P. vivax*, tremendous activity against erythrocytic stages of certain plasmodial strains, low general toxicity, and low cost, the drug also has certain undesirable qualities which augur poorly for either general or long term use. These deficiencies include slowness of action against active infections, uncertain effectiveness against diverse strains, and potentiality to induce formation of chlorguanide-resistant parasites. All these deficiencies

are especially serious where the predominating species is falciparum It must be noted, however, that chlorguanide is still being used fairly widely for routine suppression of malaria Its use for this purpose appears to be reasonably successful at a daily dose of 0 1 or 0 2 gram.

Primaquine and Other 8-aminoquinolines

The drugs dealt with up to this point owe their position in malaria therapeutics to capacities to control the growth of the erythrocytic stages of the human plasmodia; thus their use is limited to suppression of inapparent infections or to treatment of active infections The 8-aminoquinolines, although originally introduced for a similar purpose, owe their present position strictly to activity against the persisting exoerythrocytic stages of *P. vivax*, in other words, to ability to cure established infections with this plasmodium. This quality, although of little concern to persons who are permanent inhabitants of areas where malaria is endemic, is of great consequence to individuals who are only temporary residents and who on leaving these areas wish to be rid of the liability of repeated relapses

Pamaquine (known widely by its trade name *Plasmochin*) was the first of many 8-aminoquinolines introduced into medical practice. The development of this drug is an interesting chapter in malaria therapy, of which only brief mention can be made here When first made available, pamaquine was advocated as a quinine substitute to be used primarily in the treatment of active infections Its success in this area was short lived for it was soon found that doses required for successful therapy were far above ranges which could be administered safely. There were a number of reported fatalities during the early use of pamaquine and probably many more deaths which were not reported In the course of the original work, however, it was noted that pamaquine was lethal to the gametocytes of all three species of human plasmodia This effect was regarded as of particular importance in falciparum infections where sexual parasites, known to be infectious for mosquitoes, persist for some weeks after asexual stages have been cleared from the blood by administration of quinine. Subsequent to these initial observations, it was found that the gametocidal action of pamaquine could be achieved with one-quarter to one-fifth of the dose required to control erythrocytic infections As a consequence of this finding, attempts were made in certain areas to achieve more complete control of malaria through concurrent administration of pamaquine with quinine Quite unexpectedly, this work led to the finding that the combined use of pamaquine and quinine effected a significant reduction in the relapse rate in vivax malaria (59) Shortly after this, in a wholly independent investigation, it was noted that pamaquine had the capacity to prevent malaria infection when administered during the days immediately following sporo-

zoite inoculation (37). Thus in this 8-aminoquinoline there was available for the first time a drug which could prevent the establishment of malaria, which had activity against the active disease, which destroyed gametocytes, and which would prevent the relapses characteristic of *P. vivax*. Had pamaquine exhibited these qualities in doses which could be tolerated, it would have approached the qualifications of an ideal drug.

For many years these remarkable attributes of pamaquine were neither generally recognized nor understood; neither were they seriously exploited. The toxicity exhibited by the drug during initial use had created exceedingly unfavorable impressions, administration even in the small doses required for gametocidal or anti-relapse purposes provoked numerous minor reactions. These included methemoglobinemia, dizziness, nausea and intense abdominal cramping which at times made continuation of treatment difficult if not impossible. To these comparatively minor reactions had to be added the more serious complications of the severe hemolytic incidents which appeared in an unpredictable manner particularly among dark-skinned persons. The enhancement of all toxic reactions when the drug was administered together with quinacrine proved to be a further deterrent to general use With this background it is not surprising that interests in the potentialities of pamaquine were extremely limited It might be noted too that many other 8-aminoquinolines were prepared and studied during the decade which followed the introduction of pamaquine. In no instance was any of these compounds an improvement over the original drug

Interest in the 8-aminoquinolines was revived during the later phases of the malaria research program pursued in the United States during World War II. A number of factors contributed to this revival among which were: 1) new conceptions of the life cycle of the malaria parasite (cf figs 1 and 2), 2) final realization that search for drugs which exhibited only straightforward increases in schizonticidal activity was not likely to yield a compound with either prophylactic activity or ability to cure established vivax malaria, and 3) that pamaquine probably possessed a unique range of antimalarial activities even though not available in generally usable form. When systematic studies (4, 27, 40) demonstrated that pamaquine possessed true prophylactic and curative properties, a complete review of the 8-aminoquinoline field was undertaken with preparation and testing of a wide variety of new derivatives The scope of the initial effort has been described elsewhere (7, 52). It is sufficient to point out here that the programs of chemical synthesis, lower animal study, and examination in man, which were initiated during the latter years of World War II have been continued to the present day This work has shown that ability to cure relapsing vivax malaria is a general property of the 8-aminoquinolines, and has yielded a series of derivatives which are considerably more effective

and far better tolerated than pamaquine. Four compounds stand out as the best of some sixty derivatives which have been examined in vivax malaria in man or in the counterpart of this disease in the rhesus monkey These are pentaquine, isopentaquine (6-methoxy-8-(4-isopropylamino-1-methylbutylamino)-quinoline), primaquine and SN 3883 (6-methoxy-8-(4-aminobutylamino)-quinoline). Of these compounds only pentaquine is commercially available at the present writing although the most effective member of the group, primaquine, should become generally available in the near future. It seems doubtful whether either isopentaquine or SN 3883 possesses sufficient advantages to merit commercial production.

Since the pharmacological properties, and particularly the toxicity of the 8-aminoquinolines, have been such important factors in determining the usefulness of these drugs, the following comments are in order All 8-aminoquinolines studied thus far are rapidly absorbed, fixed only slightly in tissues and degraded with extreme speed. Drugs such as pentaquine and primaquine are completely eliminated from the body within 24 hours. Their elimination is accomplished largely through metabolic conversions and excretion of the degradation products in the urine. Less than 1.5 per cent of the pentaquine or primaquine which is administered can be recovered from the urine as unchanged drug (Hughes, Smith, and Schmidt, unpublished observations). As a group of compounds the 8-aminoquinolines have exhibited one of three major patterns of toxic reactions, referable to effects on 1) the central nervous system, 2) the heart and circulation, or 3) the formed elements of peripheral blood and bone marrow. Compounds which are neurotoxic usually do not exhibit other patterns of toxic reactions. Compounds which are hematotoxic may or may not have undesirable effects on the heart and circulation, and vice versa. A detailed description of these reactions and their relations to the structure of the 8-aminoquinolines has been presented elsewhere (7). Primaquine, like pamaquine, produces striking effects on the formed elements of blood, lesser effects on the heart and circulation. Pentaquine and SN 3883 on the other hand have some action on the formed elements of blood but their main effects are on the heart and circulation. Effects on blood and bone marrow components, heart, and circulation appear to be readily reversible.

The potentialities of pamaquine, as they are now understood, merit some comment, for this drug is not only the standard by which the achievements of other drugs must be measured but is still used in some areas. It is now fairly well agreed that the drug has some usefulness in preventing relapses due to established vivax infections in individuals who already possess some immunity (44). It is not highly effective in preventing relapses in non-immunes. For control of relapses it is administered for a 14-day period in daily doses of 30 mg base together with 1 or 2 grams of quinine,

the latter drug being administered to control the erythrocytic phases of the infection. Such treatment may produce severe hemolytic reactions in dark-skinned persons; if such occur, medication must be halted at once In light-skinned persons, reactions are comparatively minor and usually consist of methemoglobinemia, nausea, abdominal cramping, dizziness and headache. Even so, the above doses of pamaquine *ought to be administered* under constant medical supervision.

Pentaquine (cf. fig. 3) was the first of the newly synthesized 8-aminoquinolines which possessed activity equal to pamaquine and was less toxic Systematic studies have shown that because of lower toxicity this drug may be used to control relapses in non-immunes. When given over a 14-day period in daily doses of 60 mg base together with 1 or 2 grams of quinine, the drug has effected a high percentage of cures in non-immunes (2). Half of the above dose of pentaquine is equally effective in eradicating infections in individuals with some immunity (11), and may be employed in ambulatory cases since toxic reactions are infrequent and negligible. Doses of 60 mg. should be used only when constant medical supervision is possible. These doses frequently provoke severe abdominal cramping, nausea, dizziness, and methemoglobinemia The toxicity of both pamaquine and pentaquine makes it necessary to give these drugs in divided daily doses It should be noted that whereas both compounds possess activity against the pre-erythrocytic and erythrocytic stages of *P. vivax*, their potential use in prophylaxis, suppression, or therapy of acute attacks is vitiated by their high toxicity.

Primaquine (cf. fig 3) constitutes a marked improvement over pentaquine, both with respect to basic antimalarial activities and tolerability, and should completely replace both pamaquine and pentaquine in the control of relapsing vivax malaria Primaquine appears to be 3 to 4 times as active as the older drug when used in the same type of dosage regimen It is now well-established (Alving, Coatney, personal communications) that 14-days treatment with daily doses of 22 5 mg, together with quinine, will eradicate heavy infections with *P. vivax* in non-immunes Reasoning from experience with pamaquine and pentaquine, half of this dose, approximately 10 mg, should be equally effective against infections in immunes The tolerability of primaquine is such that these doses can be administered once daily rather than 4 or 6 hourly as is necessary with older drugs Tolerability also makes it possible to use the drug on an ambulatory basis. Problems of administration may be reduced even further by replacement of quinine with chloroquine or amodiaquin in the curative combination In this regimen chloroquine is administered during the first 3 days of the treatment period at the same dosage as is employed for simple control of erythrocytic infections

Limited observations in human malaria (63) and far more extensive studies in simian malaria (Schmidt and coworkers, unpublished observations) suggest that administration of an 8-aminoquinoline during the quiescent stage of the disease is as effective in eradicating the persisting tissue stages as treatment during the active infection. If this early evidence is supported by the results of work now in progress it would appear that usable techniques are at hand for complete control of malaria without exposure to any of the features of active disease Administration of an effective schizonticide such as chloroquine during residence in malarious regions, followed by a course of primaquine promptly upon departure from such areas should achieve this objective This end could be accomplished by using both drugs at generally tolerated levels

In concluding this discussion of the 8-aminoquinolines, attention should be drawn to early studies in both vivax and cynomolgi malaria (Alving, personal communication, Schmidt and coworkers, unpublished observations) which suggest that primaquine possesses a usable order of prophylactic activity. These same observations suggest that twice weekly administration of primaquine is sufficient to eradicate the pre-erythrocytic tissue phases. It should also be noted that primaquine possesses a usable order of activity against the erythrocytic parasites of *P vivax* although in this respect it falls far short of the accomplishments of the newer schizonticidal drugs Attention is called to these observations merely to indicate again that in one drug it is possible to achieve significant orders of activity against all developmental stages of human plasmodia

SUMMARY AND CONCLUSION

In the discussion presented above the attributes of currently available antimalarial drugs have been described in terms of their capacities to influence the various disease mechanisms involved in the human malarias The significant differences between the antimalarial activities and pharmacological properties of the newer and older compounds have been reviewed briefly It should be apparent from this discussion that whereas the ideal goal in malaria chemotherapy has not been attained in a single drug, the goal has been approached closely through combined use of several of the newer compounds The development of the 4-aminoquinolines and particularly chloroquine has made possible for the first time routine and reliable suppression of all human malarias by administration of a well-tolerated drug at widely spaced intervals These compounds likewise provide a certainty of control over active infections not found hitherto in any other drugs With the development of primaquine for the ultimate eradication of established vivax infections, mechanisms are at hand for simple and essentially complete chemotherapeutic control over all aspects of human malaria.

Assuming that economic limitations can be surmounted, the use of these chemotherapeutic measures should go far toward minimizing the ravages of malaria until such time as insect vector eradication can become a reality

BIBLIOGRAPHY

1. Adams, A R. D., Maegraith, B. B, King, J. D, Townshend, R. H., Davey, T. H and Havard, R. E · Studies on synthetic antimalarials XII. Results of a preliminary investigation of the therapeutic action of 4888 (paludrine) on acute attacks of benign tertian malaria. Ann Trop Med, 39: 225, 1945
2. Alving, A. S, Craige, B, Jr, Jones, R, Jr, Whorton, C M., Pullman, T. N and Eichelberger, L Pentaquine (SN 13,276), a therapeutic agent effective in reducing the relapse rate in vivax malaria. J. Clin. Invest., 27: 25 (part II), 1948.
3. Alving, A. S, Eichelberger, L., Craige, B, Jr., Jones, R, Jr, Whorton, C M and Pullman, T. N. *Studies on the chronic toxicity of chloroquine* (SN 7618) J. Clin Invest, 27: 60 (part II), 1948.
4. Berliner, R. W, Earle, D P, Jr, Taggart, J V, Welch, W. J, Zubrod, C. G, Knowlton, P, Atchley, J. A. and Shannon, J. A. Studies on the chemotherapy of the human malarias. VII. The antimalarial activity of pamaquine J Clin. Invest, 27: 108 (part II), 1948.
5. Berliner, R. W, Earle, D P, Jr, Taggart, J V, Zubrod, C G, Welch, W. J, Conan, N J, Bauman, E, Scudder, S T. and Shannon, J A · Studies on the chemotherapy of the human malarias VI. The physiological disposition, antimalarial activity, and toxicity of several derivatives of 4-aminoquinoline J Clin Invest., 27: 98 (part II), 1948.
6. Bishop, A and Birkett, B Acquired resistance to paludrine in *Plasmodium gallinaceum* Nature, 159: 884, 1947
7. Blanchard, K C and Schmidt, L. H Chemical Series of Potential Interest Chap III in Wiselogle's Survey of Antimalarial Drugs, 1941–1945, p 73 J W. Edwards, Ann Arbor, Michigan, 1946.
8 Carrington, H C, Crowther, A F, Davey, D. G, Levi, A A and Rose, F L A metabolite of "Paludrine" with high antimalarial activity Nature, 168: 1080, 1951
9 *Chaudhuri, R N and Rai Chaudhuri, M. N* · *Falciparum* infections refractory to paludrine. Indian J Malariology, 3: 365, 1949
10 Coatney, G R. and Cooper, W C Recrudescence and relapse in vivax malaria. Proceedings 4th International Congress on Tropical Medicine and Malaria, 629 Washington, 1948
11 Coggeshall, L T and Craige, B. Old and new plasmodicides Chap 46 in Boyd's Malariology, p 1105 W B Saunders Company, Philadelphia, 1949
12 Colonial Medical Research Committee Proguanil in prophylaxis and treatment Brit M J, 1: 585, 1949
13 Cooper, W C Summary of antimalarial drugs Pub Health Rep, 64: 717, 1949
14 Cooper W C, Coatney, G R. and Imboden, C A, Jr Studies in human [illegible]
15 C[illegible], [illegible] *mulatta.* Proc. Soc Exper. Biol & Med, 70: 360, 1949
16. Coulston, F and Robinson, F O: Artifacts and exoerthrocytic stages of *Plasmodium cynomolgi* in *Macaca mulatta* J Parasitol, 36: 28, 1950

17. COVELL, G, NICOL, W. D, SHUTE, P. G. AND MARYON, M: "Paludrine" (proguanil) in prophylaxis and treatment of malarial infections caused by a West African strain of *P. falciparum* Brit M J, 1: 88, 1949
18 CRAIGE, B, JR, WHORTON, C. M, JONES, R, JR., PULLMAN, T. N, ALVING, A S, EICHELBERGER, L. AND ROTHMAN, S.: A lichen-planus like eruption occurring during the course of chloroquine administration. J. Clin. Invest., 27: 56 (part II), 1948
19 CROUNSE, N. N.: Isolation and identification of a metabolite of chlorguanide. J. Org Chem, 16: 492, 1951.
20. CURD, F H S, DAVEY, D G AND ROSE, F. L. Studies on synthetic antimalarial drugs X Some biguanide derivatives as new types of antimalarial substances with both therapeutic and causal prophylactic activity. Ann Trop Med, 39: 225, 1945
21. EARLE, D. P., JR, BERLINER, R. W., TAGGART, J. V, WELCH, W. J, ZUBROD, C. G, WISE, N. B CHALMERS, T. C, GREIF, R. L AND SHANNON, J. A.: Studies on the chemotherapy of the human malarias. II. Method for the quantitative assay of suppressive antimalarial action in vivax malaria J. Clin. Invest, 27: 75 (part II), 1948.
22 EARLE, D. P., JR, BERLINER, R W, TAGGART, J. V., ZUBROD, C. G., WELCH, W. J, BIGELOW, F. S, KENNEDY, T. S, JR AND SHANNON, J A Studies on the chemotherapy of the human malarias. X. The suppressive antimalarial effect of paludrine. J Clin. Invest, 27: 130 (part II), 1948
23. EDESON, J F B. AND FIELD, J W.: Proguanil-resistant falciparum malaria in Malaya Brit. M J, 1: 147, 1950
24. FAIRLEY, N. H: Atebrin susceptibility of the Aitaipe-Wewak strains of *P. falciparum* and *P vivax* Trans Roy. Soc Trop Med & Hyg, 40: 229, 1946.
25 FAIRLEY, N H: Researches on paludrine (M4888) in Australia from the Land Headquarters, Medical Research Unit, Cairns Med J Australia, 1: 234, 1946
26. FAIRLEY, N. H Sidelights on malaria in man obtained by subinoculation experiments Tr Roy Soc Trop Med & Hyg, 40: 621, 1947.
27. FELDMAN, H A, PACKER, H., MURPHY, F. D. AND WATSON, R B: Pamaquine naphthoate as a prophylactic for malarial infections. J Clin Invest, 26: 77, 1947
28 FLETCHER, W. Notes on the treatment of malaria with cinchona alkaloids Study No 18 from The Institute For Medical Research, Kuala Lumpur, Federated Malay States, 1923
29 GAUD, J, SCHNEIDER, J. AND MECHALI, D.. Action comparée de la névaquine et du chloréguane en prophylaxes collectives du paludisme. Bull. Inst hyg Maroc, 9: 121, 1949
30 GEIMAN, Q The cultivation of malaria parasites Chap 9 of Most. Parasitic Infections of Man Columbia University Press, New York, 1951.
31 GRASSI, B Die Malaria Studies eines zoologen. Gustav Fischer, Jena 1901
32 HAWKING, F Activation of paludrine in vivo Nature, 159. 409, 1947.
33 HAWKING, F AND PERRY, W. L M Activation of paludrine Brit. J Pharmacol, 3: 320, 1948
34 HAWKING, F, PERRY, W. M. AND THURSTON, J. P Tissue forms of a malaria parasite (*Plasmodium cynomolgi*). Lancet, 1: 783, 1948
35 HERING, E R, PATT, H M AND LEAVITT, H J Tolerability studies of some new antimalarial drugs J. Nat Malaria Soc, 7: 322, 1948.
36 HUFF, C G. AND COULSTON, F.: The development of *Plasmodium gallinaceum* from sporozoite to erythrocytic trophozoite J. Infect Dis, 75: 231, 1944

37. James, S. P.: On the prevention of malaria with Plasmoquine. Lancet, 2: 341, 1931.

38 James, S. P., Some general results of a study of induced malaria in England Tr Roy Soc Trop Med. & Hyg., 24: 477, 1931

39. Jones, R , Jr , Pullman, T. N., Whorton, C. M , Craige, B., Jr , Alving, A S and Eichelberger, L : The therapeutic effectiveness of large doses of paludrine in acute attacks of sporozoite-induced vivax malaria (Chesson strain) J Clin. Invest , 27: 51, 1948.

40 Jones, R , Jr , Craige, B , Jr , Alving, A. S , Whorton, C M , Pullman, T N. and Eichelberger, L . A study of the prophylactic effectiveness of several 8-aminoquinolines in sporozoite-induced vivax malaria (Chesson strain) J. Clin Invest , 27: 6 (part II), 1948

41 Laveran, A Note sur un nouveau parasite trouvé dans le sang de plusiers malades atteints de fièvre palustre Bull. Acad de méd , 9: 1235, 1880

42 MacLeod, R C · Prophylactic proguanil in the southern highlands of Tanganyika Brit M. J , 1: 282, 1951.

43 Maegraith, B G , Adams, A R D., King, J. D , Townshend, R H , Davey, T. H and Havard, R E Studies on synthetic antimalarials. XIV Results of a preliminary investigation of the therapeutic action of 4888 (paludrine) on acute attacks of malignant tertian malaria Ann Trop Med , 39: 232, 1945

44. Most, H , Kane, C A , Lavietes, P. H., London, I M , Schroeder, E F and Hayman, J M Combined quinine-Plasmochin treatment of vivax malaria Effect on relapse rate Am J M Sc., 212: 550, 1946

45 Rose, F L A chemotherapeutic search in retrospect J. Chem. Soc , *2770, 1951.*

46 Ross, R. Report on the cultivation of Proteosoma, Labbe, in grey mosquitoes Indian M Gaz , 33: 401, 1898

47 Schaudinn, F Studien uber krankheitserregende Protozoen II. Plasmodium vivax, der Erreger des Tertianfiebers beim Menschen Arb. K Gesundh , 19: 169, 1903

48 Seaton, D R and Adams, A. R D · Acquired resistance to proguanil in *Plasmodium falciparum* Lancet, 2: 323, 1949

49 Seaton, D R and Lourie, E M Acquired resistance to proguanil (Paludrine) in *Plasmodium vivax* Lancet, 1: 394, 1949

50 Schmidt, L H , Genther, C S , Fradkin, R and Squires, W Development of resistance to chlorguanide (Paludrine) during treatment of infections with *Plasmodium cynomolgi* J Pharmacol & Exp Therap , 95: 382, 1949

51. Schmidt, L H , Hughes, H B and Smith, C C On the pharmacology of N_1-para-chlorophenyl-N_5-isopropylbiguanide (Paludrine) J Pharmacol & Exp Therap , 90: 233, 1947

52 Shannon, J A *Rationale underlying the clinical evaluation of antimalarial* drugs In Wiselogle Survey of Antimalarial Drugs, 1941–1945, p 177 J W Edwards, Ann Arbor, Michigan, 1946

53 Shannon, J A , Earle, D P , Jr , Berliner, R W and Taggart, J V Studies on the chemotherapy of the human malarias I Method for the quantitative assay of suppressive antimalarial action in vivax malaria J Clin Invest , 27: 66 (part II), 1948

54 Shannon, J A , Earle, D P , Jr , Brodie, B B , Taggart, J V and Berliner, R W. The pharmacological basis for the rational use of atabrine in the treatment of malaria J Pharmacol & Exp Therap , 81: 307, 1944

55 Shortt, H. E , Fairley, N H , Covell, G , Shute, P. G and Garnham, P. C. C · The pre-erythrocytic stages of *Plasmodium falciparum*. Tr. Roy. Soc. Trop. Med & Hyg., 44:405, 1951

56. Shortt, H E and Garnham, P. C. C.· Demonstration of a persisting exo-erythrocytic cycle in *Plasmodium cynomolgi* and its bearing on the production of relapses. Brit. M. J , 1: 1225, 1948

57. Shortt, H E and Garnham, P. C. C.. The pre-erythrocytic development of *Plasmodium cynomolgi* and *Plasmodium vivax* Tr. Roy Soc. Trop Med. & Hyg , 41: 785, 1948

58 Shute, P. G.. Antimalarial drugs. Brit M J , 2: 966, 1946

59 Sinton, J A , Smith, S and Pottinger, D : Studies in malaria with special reference to treatment, further researches into the treatment of chronic benign tertian malaria with Plasmoquine and quinine Indian J. M Res , 17: 793, 1929–30

60 Taggart, J V., Earle, D P , Jr , Berliner, R. W , Zubrod, C. G , Welch, W. J , Wise, N. B , Schroeder, E. F , London, I M. and Shannon, J. A.· Studies on the chemotherapy of the human malarias III. The physiological disposition and antimalarial activity of the cinchona alkaloids. J Clin. Invest , 27: 80 (part II), 1948

61 Van Goor, W. T., Lodens, J. G and Gomez, J. A · Clinical prophylaxis with proguanil and nivaquine in a community in Java. Docum neer et indones morbis trop , 2: 341, 1950

62 Warren, A J and Coggeshall, L T.· Infectivity of blood and organs in canaries after inoculation with sporozoites Am J. Hyg , 26: 1, 1937.

63 White, W. C , Cooper, W C , Coatney, G R , Culwell, W. B , Lints, H. A. and Young, M. D · Studies in human malaria. XXI The cure of St. Elizabeth strain vivax malaria by pentaquine-quinine, administered during acute attacks or during latency J Nat Malaria Soc , 7: 316, 1948

64 Williamson, J , Bertram, D. S and Lourie, E M : Effects of paludrine and other antimalarials Nature, 159: 885, 1947.

The Seasonal, Arthropod-borne, Virus Encephalitides

With Remarks on Their Relation to the General Problem of Virus Encephalitis

R. WALTER SCHLESINGER, M D.

INTRODUCTION

During the past two or three decades, our knowledge of epidemic encephalitis has undergone profound changes. While von Economo's type of the disease has virtually disappeared, at least in epidemic proportions, "new" forms have sprung up in quick succession in different places all over the world. Methods used in the rapid identification of several viruses as their causative agents and in the development of reliable and specific laboratory diagnostic tests have set a pattern for work on many other virus diseases Intensive investigation of their epidemiologic features has revealed information of far-reaching interest, not only in relation to human disease but also to infections of wild and domestic animals. Certain of the "arthropod-borne, seasonal, viral encephalitides" (41), which are the main concern of this paper, are prevalent in remote parts of the world only, and probably the majority of American physicians have never had an opportunity to see them. However, the world-wide scope of our present and future military and economic spheres of interest, coupled with ever increasing rate and volume of intercontinental travel, make greater familiarity with these diseases desirable and inevitable Even now, virus laboratories set up in various parts of the United States frequently receive and handle requests for diagnostic tests for all known forms of viral encephalitis The intelligent utilization of these diagnostic facilities can be assured only if the clinician and the laboratory worker alike have a clear picture of the position of the various viral encephalitides in the entire field of viral or non-viral infections of the central nervous system (CNS) At a time when it has become a matter of every day practice to ascribe various ill-defined disease entities to viruses, a critical survey of what we actually know about viral infections of the CNS is indicated

In this connection, it should be remembered that encephalitis, myelitis, meningitis, and the various combined terms are pathological definitions. They are applicable to all conditions in which damage to the brain, spinal cord, or meninges is associated with inflammatory reaction, regardless of whether the causative agent is infectious or noninfectious Damage to neu-

rons or to other elements of the CNS may be due either to the direct action of the noxious agent, or to some mechanical or nutritional defect, such as obstruction of blood supply, anoxia, edema, inflammatory infiltrations, or hemorrhages. The type and severity of clinical manifestations depend on the site and extent of the damage. The pathological process may be progressive or self-limiting, and the damage to specific neuronal elements may be reversible or irreversible, the clinical course varying accordingly.

The type of impairment resulting from damage to neurons depends on their specific function and is the same no matter what the nature of the damage. How can one distinguish viral encephalitides from neurological disorders of other types? What are the guiding criteria which should arouse suspicion of viral etiology? In order to bring the important issues into sharper relief, it may be instructive to review briefly the present status of our knowledge regarding viral infections of the human CNS in general. Within that broader framework, the seasonal, arthropod-borne types of encephalitis stand out as a uniform family of acute infectious diseases

GENERAL DISCUSSION

Present Status of Knowledge Concerning the Etiology of Viral Infections of the Human Central Nervous System

DISEASES IN WHICH VIRUSES HAVE BEEN ISOLATED FROM THE HUMAN CNS

The ability of a virus to cause infection of the CNS can be proved unequivocally only by its isolation from the spinal fluid or from nervous tissue of patients during the acute phase of illness or at death. Relatively few agents have ever been isolated directly in this way. The principal ones are listed in table 1. Single instances of recovery of a few other viruses from the CNS have been reported, but these findings are difficult to evaluate. In other cases, like that of measles encephalitis, confirmation is lacking because reliable laboratory tests are not available Undoubtedly many additional viruses, now only suspected of being neurotropic in man, will prove to be so once adequate methods for their isolation are developed. This applies in particular to the exanthematous diseases of childhood which, like mumps, are often accompanied by meningo-encephalitic syndromes.

Convalescence from many viral or bacterial diseases is followed on occasion by "post-infection encephalitis". Although some relationship undoubtedly exists here between the primary infections and subsequent development of neurological disorders, attempts at virus isolation from cases of post-infection encephalitis have invariably failed. The same is true for encephalopathies which occur after smallpox vaccination or other forms of immunization The pathological picture of post-infection or post-vaccina-

tion encephalitis differs significantly from that of proved primary viral encephalitides, as will be discussed below.

Among the diseases listed in table 1, rabies is the only one in which recognizable involvement of the CNS appears to be an inescapable result of infection. (As far as "B" virus is concerned, our knowledge of human infection is limited to two proved cases, both of them fatal laboratory infec-

TABLE 1

Principal Diseases in which Viruses Have Been Isolated Directly from the Human CNS

VIRUS	EPIDEMIOLOGICAL CHARACTER		PROBABLE SOURCE OF HUMAN INFECTION			MOST CHARACTERISTIC TYPE OF ILLNESS		
	Predominantly epidemic and seasonal	Predominantly sporadic, nonseasonal	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
Poliomyelitis	x		x				x	
St Louis encephalitis	x			x		x		
Western equine encephalitis	x			x		x		
Eastern equine encephalitis	x			x		x		
Venezuelan equine encephalitis	x			x		x		
Japanese (B) encephalitis	x			x		x		
Russian spring-summer enc	x			x		x		
Louping-ill	x			x		x		
Lymphogranuloma venereum		x	x			x		
Herpes simplex		x	x			x		
Mumps		x	x					x
Rabies		x			x	x		
"B" virus		x			x	x		
Lymphocytic choriomeningitis		x			x			x

tions contracted as a result of bites by spontaneously infected monkeys (103)) In infection of man with all the other viruses, clinical signs of nervous involvement are rare, although the incidence of systemic (extraneural) infection is high Thus, mumps meningo-encephalitis may occur with or without the simultaneous presence of parotitis Despite the fact that this is probably the most common and widespread of the known viral CNS infections, it is nevertheless rare in comparison with the almost universal incidence of generalized infection with mumps virus Even more striking is the discrepancy in the case of herpes with its high general infec-

tion rate and a remarkably small number of proved cases of encephalitis. Similarly, lymphogranuloma venereum encephalitis has been described only a few times, and only in patients showing primary genital lesions The other agents lead, in the majority of infected individuals, to subclinical infections or to mild, "non-specific" illnesses. It is not clearly understood whether this apparent "protection" of the CNS is due to its relative anatomical or physiological isolation, which may prevent the viruses from gaining entrance, or whether variations inherent in the virus or in host susceptibility play the chief role in determining the infected individual's fate.

Various attempts have been made to classify the viruses affecting the human CNS according to the relative degree of their neurotropism (87). Such attempts are usually based either on the type of illness most frequently seen in man or on the more or less selectively neurotropic behavior of the viruses in laboratory animals That the latter is an arbitrary criterion is evident from the fact that mumps virus has shown no neurotropic tendencies under experimental conditions On the other hand, certain viruses which rarely, if ever, produce neurological complications in man, have been found to proliferate preferably or exclusively in nervous tissues of laboratory hosts to which they were adapted Among these are yellow fever (135), dengue (110, 115), Rift Valley fever (74), and Colorado tick fever (67) viruses Thus, neurotropism in experimental animals does not justify the conclusion that a virus attacks the human CNS, even if it is capable of infecting human beings.

Neither is the type of illness produced most frequently in man a valid measure of an inherent degree of neurotropism: it has already been mentioned that the incidence of CNS involvement in persons infected with the viruses listed in table 1 is low This conclusion is based (a) on the known high prevalence of mumps, lymphogranuloma venereum, and herpes simplex, diseases which can be diagnosed readily in the absence of neurological complications, and (b) in the case of poliomyelitis and the arthropod-borne encephalitides, on the findings of antibodies in the sera of a large proportion of the "normal" population living in areas in which the viruses are prevalent The viruses of the poliomyelitis group which, until recently, were considered to be "strictly" neurotropic can no longer be set apart from the other agents on that basis The frequent demonstration of poliomyelitis virus in the feces of persons with or without paralysis suggests that it may be capable of multiplying outside of the CNS This is borne out by the recent success of Enders and his group (30) and of others in the propagation of poliomyelitis viruses *in vitro* in minced human or monkey non-neural tissues It would appear that the main difference between "non-neurotropic" viruses, such as mumps, herpes, or lymphogranuloma inguinale,

and those ordinarily classified as neurotropic, is that the former cause a well-defined extraneural disease, while the systemic phase of infections with the latter is "non-specific" or latent in nature.

Separation of the different forms according to clinical characteristics is suggested by the arrangement in table 1. The most *characteristic* type of clinical syndrome need not be the most *frequent* one. Thus, the incidence of "aseptic meningitis" due to poliomyelitis virus may exceed that of the myelitic (paralytic) form. It is quite possible that more general use of diagnostic tests with herpes simplex antigens may reveal a similar situation for this virus, as suggested by recent studies of Afzelius-Alm (2). Despite such reservations, clinical characterization, combined with epidemiological and anamnestic information, is helpful in directing the differential-diagnostic leads to one or the other of the groups as they are listed in table 1

This list reflects the present status of laboratory work in this field. Its inadequacy in relation to the overall incidence of neurological disorders of probable viral etiology will become more evident in a later section dealing with the efficacy of specific laboratory diagnostic procedures under varying circumstances. The incompleteness is particularly emphasized by the rapidity with which agents are being added to the list of suspected causes of CNS infection. Three of the most recent additions deserve special mention.

MENGO ENCEPHALOMYELITIS

In 1946, a virus was isolated from the blood of a laboratory worker in the Yellow Fever Research Institute, Entebbe, Uganda, in Africa. This patient was suffering from a short illness suggestive of encephalitis, and it was shown that serum obtained on the second day of his illness contained no antibody, while a sample taken in convalescence did neutralize the new virus (Mengo encephalomyelitis virus) (24). Subsequently, this virus was found to be indistinguishable (23, 139) from a number of recently isolated strains of varied origins, viz (a) encephalomyocarditis virus originally recovered from anthropoid apes (53), (b) "MM" (61) and Columbia "SK" (60) viruses which are believed to have originated from wild or laboratory rodents. Warren et al (138) found antibody neutralizing these agents in the sera of wild rats trapped in various areas of the United States. The relationship of this group to infection of the human CNS is suggested not only by the case from which Mengo virus was obtained but also by the finding of antibodies in the sera of convalescents from "aseptic meningitis" or other neurological syndromes in the Philippines (121) or elsewhere (36, 62).

COXSACKIE *VIRUSES*

Since 1948, much work has been done on a group of viruses which have been isolated from the stools of patients suffering from "poliomyelitis-

like" illnesses, and which are characterized by their ability to cause fatal myositis and fat-necrosis in infant mice (20). Serological surveys have shown that these viruses are widely distributed in different parts of the world, and suggest that they may cause "aseptic meningitis" or even paralytic disease However, at the present time their true significance in relation to infections of the human CNS is somewhat obscured by the recent simultaneous finding of poliomyelitis and Coxsackie viruses in stools of the same patients (3), and of concomitant rise in specific antibodies against both viruses during convalescence (80). On the other hand, there is clear-cut evidence in the form of accidental infection in several laboratory workers with Coxsackie viruses which indicates that they are pathogenic for man (118). The problem which requires further clarification is their role in primary infections of the CNS. This question awaits the isolation of the agent from the CNS of patients

DURAND'S DISEASE

Durand (28) has described a febrile illness from which he was suffering and which he transmitted to two human beings by experimental inoculation of his blood. The virus held responsible for this illness was pathogenic for guinea pigs Cerebrospinal fluid obtained from one of the experimental subjects six days after inoculation contained 65 cells per cmm and was infectious for a guinea pig Despite these findings, the patient showed no neurological signs. Subsequently, it was shown by Findlay (33) that this virus is capable of causing laboratory infection in man He found no relationship of the agent to a variety of other viruses. Its significance as a possible cause of spontaneous (i e., outside of the laboratory) CNS infection of man has not been established.

Several additional viruses were newly isolated in recent years as by-products of yellow fever or encephalitis survey studies in Africa, South America, or California. These bear certain relationships to the seasonal encephalitis viruses, but their role in infection of the human CNS is entirely problematical They will be discussed in greater detail in the next section

Even if one considers only viruses of proved ability to infect the human CNS, one is impressed with the susceptibility of this organ to such a heterogeneous group of agents Some of them, notably poliomyelitis and sporadic forms of encephalitis, have been known for a long time and have occurred in all parts of the world. The different types of "arthropod-borne encephalitis", on the other hand, are all "new" diseases Each one of them is prevalent only in limited parts of the world They have much in common, even though the different viruses are readily distinguishable. Their impact as disease-producing agents is revealed by the recent history of epidemic encephalitis.

and those ordinarily classified as neurotropic, is that the former cause a well-defined extraneural disease, while the systemic phase of infections with the latter is "non-specific" or latent in nature.

Separation of the different forms according to clinical characteristics is suggested by the arrangement in table 1. The most *characteristic* type of clinical syndrome need not be the most *frequent* one. Thus, the incidence of "aseptic meningitis" due to poliomyelitis virus may exceed that of the myelitic (paralytic) form. It is quite possible that more general use of diagnostic tests with herpes simplex antigens may reveal a similar situation for this virus, as suggested by recent studies of Afzelius-Alm (2). Despite such reservations, clinical characterization, combined with epidemiological and anamnestic information, is helpful in directing the differential-diagnostic leads to one or the other of the groups as they are listed in table 1.

This list reflects the present status of laboratory work in this field. Its inadequacy in relation to the overall incidence of neurological disorders of probable viral etiology will become more evident in a later section dealing with the efficacy of specific laboratory diagnostic procedures under varying circumstances. The incompleteness is particularly emphasized by the rapidity with which agents are being added to the list of suspected causes of CNS infection Three of the most recent additions deserve special mention.

MENGO ENCEPHALOMYELITIS

In 1946, a virus was isolated from the blood of a laboratory worker in the Yellow Fever Research Institute, Entebbe, Uganda, in Africa This patient was suffering from a short illness suggestive of encephalitis, and it was shown that serum obtained on the second day of his illness contained no antibody, while a sample taken in convalescence did neutralize the new virus (Mengo encephalomyelitis virus) (24). Subsequently, this virus was found to be indistinguishable (23, 139) from a number of recently isolated strains of varied origins, viz. (a) encephalomyocarditis virus originally recovered from anthropoid apes (53), (b) "MM" (61) and Columbia "SK" (60) viruses which are believed to have originated from wild or laboratory rodents. Warren et al (138) found antibody neutralizing these agents in the sera of wild rats trapped in various areas of the United States. The relationship of this group to infection of the human CNS is suggested not only by the case from which Mengo virus was obtained but also by the finding of antibodies in the sera of convalescents from "aseptic meningitis" or other neurological syndromes in the Philippines (121) or elsewhere (36, 62).

COXSACKIE VIRUSES

Since 1948, much work has been done on a group of viruses which have been isolated from the stools of patients suffering from "poliomyelitis-

clinical and epidemiological characteristics as well as to the laboratory procedures leading to their identification

JAPANESE (TYPE B) ENCEPHALITIS

During the summer of 1924, Japan was ravaged by an explosive epidemic of encephalitis similar to and possibly related to "Australian X disease". The total number of cases reported for that summer exceeded 6,000 with a mortality rate of about 60 per cent. Japanese authors reported that smaller outbreaks of a similar illness had been observed since 1871. They believed that it differed, clinically and epidemiologically, from von Economo's type which had been seen in Japan (64).* The evidence for clinical distinctness, however, was not sharply defined or generally accepted. In 1929, several years before the specific etiology of the Japanese summer encephalitis was established, the Matheson Commission issued its first Report on the Etiology, Epidemiology, and Treatment of Epidemic Encephalitis All the published data available at that time were reviewed critically by the authors against a background of experience with von Economo's disease in the United States and in Europe. The following conclusion (p. 380) is of particular interest: "Although the Japanese authors quoted conclude that Encephalitis B is a special form of encephalitis closely related to Encephalitis A, *it does not appear that there are satisfactory grounds for making a differentiation, either between these two types in Japan, or between the Japanese and the European or American form of the disease*". Since both forms of the disease presented variable clinical manifestations with a high degree of overlapping, this skepticism was not surprising It was only in retrospect, after the etiology of Japanese encephalitis was identified, that the distinguishing clinical features became more clearly apparent.

Several Japanese investigators reported suggestive results of attempts at isolation of the causative agent, but it was not until 1934 that the virus of Japanese encephalitis was accepted as a separate entity (50, 65, 140). Since then, numerous strains of identical immunological and biological character have been isolated during outbreaks occurring in Japan, China, Korea, and Okinawa. Varying numbers of cases were reported in Japan proper every summer from 1924 to 1945, with a total of about 26,000 cases. Since the beginning of military operations by the United States in endemic areas, Americans have been affected in several outbreaks of the disease on Okinawa

* The two forms were designated as "Type A" (Economo) and "Type B" (summer epidemic), and the virus subsequently isolated from the latter type is usually referred to as Japanese Type B en[illegible]
this nomenclature be replaced [illegible]
cially since the term "Type A" [illegible]
disease This suggestion will be followed in the present chapter.

THE SEASONAL, ARTHROPOD-BORNE ENCEPHALITIDES

Historical Review

Well-documented history of epidemic encephalitis has its beginning with the disastrous and world-wide outbreak of von Economo's disease, prevalent between 1916 and 1923. Its etiology remains unknown despite intensive efforts of many investigators who had at their disposal an abundance of clinical material. This failure of many attempts to determine its cause, with some degree of consistency, is indeed one of the distinguishing characteristics which set von Economo's encephalitis apart from other forms which have taken its place. Since 1923, von Economo's disease has not recurred in epidemic proportions Occasionally, one sees sporadic cases which fit its original description and which cannot be attributed to any of the known encephalitogenic agents. The alarming threat posed by encephalitis at the end of the first World War undoubtedly alerted the medical profession to the danger and stimulated the profitable investigation of the subsequent episodes

AUSTRALIAN X DISEASE

Historically, the first large-scale encounter with a form of epidemic encephalitis believed to be distinct from von Economo's disease was a series of epidemics occurring in 1917 and 1918 in various parts of Australia A report on this disease (19) was based on detailed studies of 134 cases of which 94 were fatal Although the symptoms and the clinical course had some features deviating from the Economo type, the most striking difference lay in their seasonal incidence Epidemics of von Economo's disease occurred characteristically during winter and early spring, while Australian X disease was limited to the summer This suggested a basic difference in the epidemiology of the two types The Australian workers succeeded in transmitting the disease from three fatal human cases to *rhesus* monkeys by means of intracerebral inoculation of brain and cord tissue The causative agent appeared to be a virus It was said to be pathogenic not only for monkeys but also for sheep, calves and horses. Subsequently, its virulence was lost on storage and the disease has not reappeared in Australia since 1926 (91).* Therefore, its relationship to subsequent outbreaks in other parts of the world has never been clearly established Nevertheless, this episode set a pattern which the later ones followed closely with regard to

* Recent reports from Australia indicate that an outbreak of encephalitis in Murray Valley, Australia, in 1951 had been identified as being due to a virus closely related that of Japanese encephalitis This would seem to strengthen the prevailing opinion that Australian X disease may have been caused by the Japanese virus (see Editorial "Murray Valley Encephalitis", Med J. Australia, 38: 526–527, 1951)

Outbreaks of this disease were limited to the period from May to August. The mortality rate was about 30 per cent Later, similar forms of encephalitis were reported from Siberia and European Russia, but with much lower mortality rates. There is some question, however, whether the latter outbreaks were caused by the Far Eastern spring-summer encephalitis virus or by the closely related louping-ill virus. The total number of cases caused by these two viruses in epidemics in Russia could not be ascertained, but the amount of work done by several teams of Russian investigators indicates that in endemic regions, encephalitis was a public health problem of major importance (119, 132)

LOUPING ILL

Louping ill is a tick-transmitted form of virus encephalitis of sheep (93), enzootic in Scotland for more than a century. Interest in this agent as a possible cause of human encephalitis dates back to laboratory infections first reported in 1934 by Rivers and Schwentker (99) There has been some speculation as to whether the relatively benign form of encephalitis of European Russia may be due to louping-ill rather than to Russian spring-summer encephalitis virus, inasmuch as a close immunological relationship exists between these two agents (18). This possibility has found support in accounts of outbreaks of human encephalitis due to louping-ill virus in Czechoslovakia (92). Recently, two cases of human infection with this virus have been seen in England, and they furnish definite evidence of the pathogenicity of the sheep virus for man (21).

Thus, the history of human epidemic encephalitis since the end of the first World War has been highlighted by several outbreaks of previously unrecognized forms. Despite their geographic separation and their immunologic distinctness, the viruses share certain properties which establish them as a "new" family of encephalitogenic viruses Aside from their proved ability to cause encephalitis in man, they have in common (a) infectivity for a wide range of natural and experimental host species, (b) pronounced seasonal incidence; (c) much direct and indirect evidence for their transmission in nature by biting arthropods. Knowledge of these viruses and the diseases they cause has developed rapidly. Infection of the human CNS with equine encephalomyelitis viruses was not recognized until five and seven years, respectively, after their original isolation from horses With this fact in mind, it is perhaps justifiable to complete this historical review with brief reference to a few recently isolated viruses which have much in common with this group, although their relationship to human encephalitis is as yet unknown

SUSPECTED ADDITIONAL AGENTS

Isolation of the viruses listed in table 2 may be considered as more or less accidental by-products of epidemiological field studies on yellow fever

(136), in Korea (111), and in Japan (144). The virus is widely distributed in the areas adjacent to the Sea of Japan and the Yellow Sea. In 1948, it was responsible for an outbreak on Guam (44).

While knowledge on this form of encephalitis accumulated, seasonal epidemics of "summer encephalitis" erupted in several other parts of the world.

ST. LOUIS ENCEPHALITIS

In 1933, a sharply localized outbreak of encephalitis occurred in St. Louis and Kansas City and the surrounding area. The number of cases was approximately 1,400 (77). The similarity of the disease to the Japanese form was immediately apparent. The virus of St. Louis encephalitis was transmitted to monkeys (85) and to mice (142), and found to be immunologically distinct from Japanese encephalitis virus (143). Smaller epidemics caused by this virus occurred in 1934 and 1937. Despite the localized and fleeting character of these outbreaks, evidence has accumulated for much wider distribution of the virus throughout the Central and Western United States and for endemicity in certain sections

EASTERN, WESTERN AND VENEZUELAN E E

Since 1938, it has been apparent that the Eastern and Western types of equine encephalomyelitis virus are capable of causing encephalitis in man. These two viruses have been responsible for several serious epidemics in various sections of the United States and Canada. The virus of Eastern E.E was first isolated from diseased horses by TenBroeck and Merrill (134). The only major outbreak of human encephalitis due to this virus occurred in 1938 in Massachusetts (32) Thirty-four cases were recognized, with a mortality rate of 65 per cent. In the same year, the virus of Western E E, first isolated from horses in 1931 by Meyer, Haring and Howitt (81), was recovered from human brain tissue (54) Three years later, an epidemic caused by this virus affected about 3,000 people in North Dakota, Minnesota, Manitoba, Saskatchewan, and adjacent provinces of Canada (26, 29, 70). The virus has been endemic in certain regions of California where smaller epidemics and sporadic cases have occurred frequently during the past few years.

A third type of equine encephalomyelitis, the Venezuelan, has been responsible for two fatal human cases on Trinidad, B.W.I. (96), and is known to have caused several mild infections in laboratory workers, both in South America and in the United States

RUSSIAN SPRING-SUMMER (TICK-BORNE) ENCEPHALITIS

In 1937, a distinct virus was isolated from cases of encephalitis occurring in the far eastern regions of the Soviet Union, chiefly among forest workers

Clinical Features

GENERAL REMARKS

Virus isolation and serological tests have been used only in recent years on a fairly large scale in the diagnosis of arthropod-borne encephalitides Application of such specific diagnostic methods has facilitated the delineation of the clinical characteristics of these maladies The clinical picture of viral encephalitis is highly variable, and many other illnesses can simulate either part or all of its signs and symptoms In any epidemic, or in endemic areas, the danger exists that unrelated cases are labeled as the disease which is anticipated or known to occur. In the case of the arthropod-borne encephalitides this tendency is all the more common, since the season of highest prevalence coincides with that of other arthropod-borne diseases and, especially, of poliomyelitis It is not surprising, therefore, that in outbreaks in which specific diagnostic tests were applied, the experience has been time and again that the number of clinically diagnosed cases far exceeded those in which the specific cause was verified in the laboratory (cf Sabin's report (101) on Japanese encephalitis in American troops on Okinawa in 1945: of 38 cases whose illness justified the suspicion of a viral infection of the CNS, only 12 could be shown to have Japanese encephalitis). It seems almost certain that under ordinary circumstances the majority of these cases would have been reported as part of the same outbreak (See below under specific diagnostic methods) Undoubtedly much of what would be said of the clinical aspects of summer encephalitis would be misleading if the description were based on unreserved acceptance of the many reports on unconfirmed epidemics On the other hand, if it were limited to consideration of the relatively few etiologically confirmed cases, the true variability of the diseases would probably not become clear

The best way out of this quandary is to base the description of essential clinical features on the findings in large and well-studied outbreaks which were so sharply limited geographically and temporally that the simultaneous presence in epidemic proportions of unrelated illnesses could not have escaped attention One episode which fulfilled these requirements was the St Louis encephalitis epidemic of 1933 Excellent clinical reports on this outbreak have been published by Hempelmann in Public Health Bulletin No 214 (94) The similarities between St. Louis encephalitis and encephalitis caused by the other arthropod-borne viruses are so striking that it will serve here as a model for the other forms.

GENERAL SYMPTOMATOLOGY

St Louis Encephalitis as a Model (Hempelmann's Description)

The clinical description by Hempelmann was based on careful study of 786 patients hospitalized during the 1933 epidemic. 106 of these were chil-

in Africa and Brazil and on encephalitis in California, in the course of which attempts at virus recovery from suspicious human cases and from lots of trapped mosquitoes were made routinely. All these viruses are highly pathogenic for mice after intracerebral inoculation. Serological evidence for infection of human beings has been found in people living in the regions in which they were discovered. In the case of West Nile virus, some immunological relationship to Japanese and St. Louis encephalitis viruses has been

TABLE 2

Viruses Which Are Not Known to Have Caused Encephalitis in Man in Nature But Are Otherwise Similar to Known Encephalitogenic Viruses

NAME OF VIRUS	REGION OF PREVALENCE	YEAR OF FIRST ISOLATION	DONOR SPECIES	EVIDENCE OF PATHOGENICITY FOR MAN
West Nile (129)	Uganda	1937	Man (blood)	a Febrile illness of donor b Antibody in "normal" persons in area of origin
Bwamba fever (130)	Uganda	1937–38	Man (blood)	Febrile illness in 9 donors
Semliki forest (126)	Uganda	1942	Mosquito	Antibody in "normal" persons in area of origin
Bunyamwera (128)	Uganda	1943	Mosquito	Antibody in "normal" persons in area of origin
Ntaya (127)	Uganda	1943	Mosquito	Antibody in "normal" persons in area of origin
Uganda S (25)	Uganda	1947	Mosquito	Antibody in "normal" persons in area of origin
BFS-283 (49)	Calif	1944	Mosquito	a Antibody in 1 child with encephalitis b Antibody in "normal" persons in area of origin
Ilheus enc (69)	Brazil	1944	Mosquito	Antibody in "normal" persons in area of origin

reported (72, 125), thus strengthening the case for including at least this virus in the group of encephalitogenic agents This argument is weakened by the fact that similar serologic relationship has been found by Sabin (104) for West Nile, Japanese encephalitis, yellow fever, and dengue viruses. The last two, although neurotropic after adaptation to mice, cannot be classified among the primary causes of human encephalitis In relation to the human encephalitis problem, then, the list of agents in table 2 is a tentative projection into the future· any one of them might follow the chronological pattern described for the equine encephalitis viruses There is no reason to assume that the roster of encephalitis-producing viruses is complete.

fection. The *disease is ushered in abruptly with headache and fever*, and without prodromal manifestations of any sort Less frequently, the onset is gradual. The temperature is ordinarily high, at least 102° or 103°F , and at times 105° or 106°F., and nausea or vomiting are frequent Chills or chilly sensations, lassitude, weakness, myalgias and abdominal pain may occur, but are less frequent than in the first group discussed Occasionally, especially in children, *convulsions* may take the place of chills In general, *neck rigidity*, often accompanied or even preceded by *stiffness of the spine* below the cervical region, was perhaps the commonest objective finding In the majority of instances, the *Kernig* sign was positive at some time during the course of the disease, but not always in the early phase In some cases, however, despite marked head retraction, the Kernig remained absent throughout the entire illness As regards the other *reflexes, the most constant finding was an absence of the abdominals*, even in the first few days of the illness Occasionally, however, especially in children, they remained unchanged despite other obvious evidence of serious invasion of the central nervous system. The *knee jerks* and other *tendon reflexes* were *inconstant*, being exaggerated oftener than diminished. The Oppenheim, Gordon, Chaddock, and Babinski tests varied not only in different patients, but at different times in the same patient As a rule, no reliance could be placed on abnormal plantar reflexes or ankle clonus in arriving at a diagnosis of encephalitis, although almost all combinations of such pathological signs were observed. The pupils, as a rule, were small, but equal, and reacted to accommodation Reaction to light was occasionally sluggish

"Coincidently with the development of the above-mentioned symptoms, or at least within one or two days of the onset of the illness, many patients showed definite evidences of *mental impairment* Some were completely disoriented as to time and place, others remembered their own names and ages, but could not recall where they lived, many during convalescence had no recollection of lumbar punctures, the early part of their illness, or how they had come to the hospital Despite the popular name of 'sleeping sickness', drowsiness was by no means always present In fact, *excitement* or mild *delirium* were not uncommon, and a few patients actually suffered from *insomnia*. Nevertheless, the majority of individuals with moderately severe encephalitis showed a tendency to *drowsiness or mental apathy*, and some went into real *coma* Rarely, however, was the drowsiness so pronounced that the patient could not be aroused at least momentarily to answer a few simple questions, following which he usually lapsed again into a peaceful sleep At the height of the symptoms, there was often great *difficulty in speaking*, ranging from complete aphasia in a few, to thick or slurred speech, running words together, etc Unquestionably, in certain instances such speech difficulty was due, at least in part, to tremors of the lips and tongue, but true aphasia occurred as well as dysarthria

mild strabismus were observed, usually disappearing within a few days Complaints of slightly blurred vision early in the disease were considerably more frequent, but ordinarily lasted only for a day or two Ptosis was so rare that its presence was noted only a few times in this tabulation of 786 cases, and then it was of mild degree and persisted only a few days . *Nystagmus*, both lateral and vertical, was observed in a few cases, but disappeared within a few days. *Ophthalmoscopic* examinations by competent ophthalmologists failed to reveal any striking abnormality of the fundi

" . . *Vertigo* occurred in about one-fourth of the cases, almost always at the onset

dren under 16 years of age. The following quotations were taken from the Public Health Bulletin No 214, a monograph on the St. Louis outbreak published in 1935 by the U. S. Public Health Service.

"Even a brief study of the records reveals that there was a wide variation in the clinical picture observed, especially as regards intensity of the infection, general *constitutional response to its presence*, and the reaction of the nervous system Further consideration, however, suggests that there are certain basic symptoms which, from their frequency, seem especially characteristic of the disease, and about which may be built up a complete picture of the affection as observed during the 1933 epidemic With an understanding of such main features of the condition, variations in symptomatology can usually be explained by differences in the severity of the infection, or degree of involvement and localization of lesions in the central nervous system. Moreover, in many instances, particularly in the aged, existing pathology at the time of the onset of the illness so modified the clinical aspects as to materially increase the difficulty of diagnosis.

"The clinical symptoms which came to be looked for as especially significant of this disease may be enumerated as follows· *Abrupt onset of fever; headache; nausea or vomiting, mental confusion or disorientation; tremors* of hands, tongue, or lips, *difficulty in speech, drowsiness, stiff neck or spine;* and positive *Kernig* and *Brudzinski* signs Lumbar puncture in such a suspected case was relied upon to verify the diagnosis by *revealing clear spinal fluid with increased mononuclear cell count and no diminution in sugar* content In the main, the significant symptoms are referable to the lesions of the central nervous system, but in addition there are others which seem rather to be called forth by the toxemia or systemic reaction to a more or less general infection"

Prodromal Stage "In the latter group may be placed, for example, such complaints as pronounced malaise, extreme lassitude, chills or chilly sensations, grippy aches in back or limbs, nausea and abdominal pains, and in occasional instances, slight conjunctivitis with photophobia, sore throat, or other signs of a mild upper respiratory infection Accompanying these manifestations, there were usually fever and a certain amount of headache Physical examination, however, reveals no abnormality indicative of participation of the brain or meninges in the infection, and hence it is not surprising to find a tentative diagnosis of grippe or influenza made at this time In occasional instances, even at this early stage, patients will complain of pain in the neck muscles, and a more careful examination may arouse some suspicion of slight cervical rigidity In the type of case without neurological symptoms at the start, *this stage of invasion*, if it may be called such, lasts from one to four days, or even longer, and during this time not infrequently a marked amelioration of symptoms occurs, the temperature ranging lower and a headache subsiding Suddenly, however, the temperature again rises abruptly to 103°, 104°, or even 105°F , the [illegible] ...alitis develops, [illegible] it on, the clin- [illegible] ic type of dis- [illegible] f the so-called "two-humped type" of fever in certain cases of acute poliomyelitis The number of patients exhibiting prodromata of this sort, however, is apparently distinctly smaller than the number belonging in the second group in which such symptoms are either absent or so mild as to be entirely overlooked"

Acute Stage "A *more usual* type of the disease, then, is that in which the characteristic encephalitic or meningeal symptoms appear at the very outset of the in-

onset of the disease, and nausea, chilly sensations, or extreme lassitude at the same period Moreover, the *majority* of such patients had some suggestion of *spinal or neck rigidity* in the early phases, many had *absent abdominal reflexes*, and a few showed other abnormalities such as changes in the knee jerks and plantar reflexes With such vague and indefinite symptoms, it is not surprising to find a diagnosis of malaria or typhoid fever made, especially in those cases having an associated leucopenia Disorientation and other mental symptoms, however, were almost always entirely absent, and drowsiness was infrequent and not pronounced when present The temperature, which rose to 102° or 103° in the early stages, had usually returned to normal within five to seven days, with coincident improvement in all the symptoms. The spinal fluid, even in such mild cases, usually revealed the typical increase in mononuclear cells and helped establish the diagnosis . "

Certain Features of the Acute Phase of Other Arthropod-Borne Encephalitides

The variability of the symptoms of St Louis encephalitis, so apparent from Hempelmann's description, is equalled by that of other types of encephalitis. More than that, the relative frequency of individual signs and symptoms, and the average severity of cases differ as much from one to another epidemic of the same type as between different types. Aside from the manifestations mentioned, there are a few features which have been more striking in some outbreaks than in others. Among these are the following:

Japanese Encephalitis. Recent American observers believe that infection with this virus has an all-or-none effect, i e , that in this form the syndrome of "aseptic meningitis" does not, or only rarely, occur. It is claimed that the infection is either entirely latent, or that CNS involvement is accompanied by some manifestation of cerebral damage (101, 136) It is perhaps significant that among the few confirmed cases which more or less fit into the category of benign meningitis there were at least two American soldiers who had been partially immunized (case 12 of Sabin's Okinawa series (101) and case 4 in Wyatt's report (141)) Paralyses, usually shifting from flaccid to spastic, have been seen more frequently in severe native cases (73), especially in children, than in Americans (101) in the same outbreak In most cases, pareses are transient but they may persist in severe cases Rigidity, incoordination, purposeless (athetoid) movements, or tremors are common. Sabin has pointed to the significance of absent cremasteric reflexes and has emphasized the frequency of bradycardia during the febrile period.

Russian Spring-Summer Encephalitis. In this type, the incidence of paralysis is higher than in the other forms According to Silber and Soloviev (119), ascending paralysis of the Landry type with eventual bulbar involvement is common, and flaccid paralysis of the upper extremities with subsequent atrophy of the brachial and cervical muscles (neck drop) occurs in about 20 per cent

of the disease and disappearing within a day or two. The dizziness as a rule was so mild that its presence was revealed only upon definite inquiry. A very few instances of real ataxia suggesting a cerebellar origin were observed, but these, too, cleared up rapidly.

"*Fever* in the typical encephalitis case was highest in the first two or three days of the infection and then, in most instances, fell by rapid lysis, so that the normal was reached within a week or ten days of the onset. Somewhat less frequently, the temperature fell by crisis, and in general, it was uncommon to have any subsequent exacerbation of the fever except when complications developed Occasionally, however, the febrile course, even in uncomplicated cases, was very much more prolonged, even up to a month or six weeks As a rule, the patient's general condition improved as the temperature returned to normal, but exceptions were observed in which the fever disappeared although the patient remained confused and disoriented, or had tremors even as long as three or four weeks later. The *pulse* was ordinarily proportional to the temperature, but it was not at all infrequent to find a marked *bradycardia*, and less commonly, a tachycardia The blood pressure was unchanged except in those individuals with preexisting arteriosclerosis, nephritis, or cardiac conditions.

"Mention has been made of the frequency with which nausea and vomiting occurred in the early stages of the disease It should be emphasized, however, that this was rarely of the forceful type seen in meningitis or other conditions in which there is associated a marked increase in intracranial pressure. As might be expected under the circumstances, the spinal fluid in most cases was not under greatly increased pressure, and reaccumulated slowly after lumbar puncture Constipation was very common, probably due in part to reduction in the ingestion of solid food, but a small number of patients had severe diarrhea, occasionally with a considerable amount of blood in the stools Retention of urine was common and in some instances *incontinence of feces and urine* occurred, the latter at times as an overflow phenomenon when the bladder was distended

"It is important, too, to record the fact that in a few cases *paralyses* developed during the course of the acute illness, before the febrile period had ended When these involved the extremities, monoplegias, hemiplegias and diplegias were observed, usually of the *spastic* type Facial paralysis was occasionally a somewhat later complication, occurring toward the end of the second week As a rule, however, all such manifestations had disappeared by the end of the third or fourth week of the disease Despite this generally favorable course, in a few cases there was *marked mental deterioration* and *spasticity of all four extremities* persisting long after the usual isolation period of three weeks had passed At the first follow-up examination, [illegible]

Mild Forms—'Aseptic Meningitis' "Finally, in discussing the clinical types of encephalitis as observed in the 1933 epidemic, it is important to mention a third group, made up of *mild or abortive* cases, in which the symptoms were so mild and [illegible]

encephalitis and diagnostic [illegible] It seems probable that more [illegible] of this type were overlooked than were discovered As a rule, however, careful examination and history revealed at least some suggestive symptoms, as, for example, slight tremors of tongue and hands, mild transient vertigo or blurring of vision at the

defervescence, the outlook for survival or complete recovery is dim. Gradual deterioration of all vital functions may continue for months or years.

The extent of persistent residua depends on the degree of irreversible damage to nervous tissue Residua in some of the severe cases of Eastern E E, seen in 1938, are illustrative. One child (R. R.), a month old at time of onset, presented a picture of a "decerebrate animal" eleven months later (37). Serious damage to the brain was evident from the encephalogram taken four months after onset (31). This child finally died in 1944, having

TABLE 3

Time of Death in 201 Fatal Cases, 1933 Encephalitis, St Louis Area

INTERVAL BETWEEN ONSET AND DEATH	NUMBER OF CASES
1 day	2
2 days	10
3 days	11
4 days	23
5 days	24
6 days	25
7 days	16
Total 1st week	111
1–2 weeks	62
2–3 weeks	10
3–4 weeks	8
4–5 weeks	5
5–12 weeks	5
Total	201

From Public Health Bulletin No 214, 1935, p 23

had as many as 42 grand mal and 31 petit mal seizures per month (4). Another child in the same series (M C.) had internal hydrocephalus 4½ months after onset and after one year had mental deficiency, right hemiplegia, impaired vision, and other serious disorders. At the age of 9½ years, eight years after onset, her mental age was one year, she presented serious behavior problems, and remained spastic-hemiplegic on the right side, with periodic generalized convulsions. Similar instances have been reported for the other types of encephalitis. Figure 1 illustrates severe residua in a child convalescent from Japanese encephalitis The patient was a five-year old Okinawan native who was discharged from the hospital about two months after onset (73), at which time he was described as "of the mental level of an idiot, he takes food, cries out in pain, moans when

Louping Ill and Venezuelan E.E. Few reports on these diseases occurring in epidemic form have appeared; not enough to say whether they present any distinguishing features. The few cases which have been reported fit into the description of the other forms given above.

Encephalitis in Infants and Children

In infants and children, encephalitis caused by Eastern E E. (31), Western E E. (79), St. Louis E. (94), and Japanese E. (73), is characterized by the high incidence of convulsions. Especially in the very young, there is often other evidence for marked damage to the cerebrum in the form of general rigidity, spastic paralysis, tremors and bulging fontanels.

INCUBATION PERIOD

Estimates of the incubation period of the various forms of arthropod-borne encephalitis range from 4 to 14 days, and this appears to be reasonable by analogy to findings in experimental animals.

DURATION

In the St Louis outbreak, as in epidemics of other forms of summer encephalitis, the duration of the illness was as variable as the symptoms Mild or abortive cases may last from two to three days.

When the illness is of average severity, it lasts one to three weeks. In the more severe cases, the acute phase may progress directly into a protracted subacute or chronic stage which may go on for many weeks or even months. In most cases, residual symptoms and signs improve gradually during the subacute or chronic stage.

PROGNOSIS, RESIDUA, SEQUELAE

The ultimate outcome of encephalitis due to one of the arthropod-borne viruses is difficult to predict during the first stormy days of the acute phase Even patients who had been comatose for several days or who had serious mental or motor impairments have been known to recover completely Death in uncomplicated encephalitis usually occurs during the first ten days of illness. This is characteristic of all forms. Table 3 gives the time of death in 201 fatal cases in the St. Louis outbreak of 1933 Often, defervescence is accompanied by dramatic improvement of the systemic and neurologic signs, and this is a favorable omen. Recovery of normal function, however, may proceed very slowly Impairment of mental capacity or personality changes may persist for many months with ultimate return to normal. General rigidity and spastic pareses of the extremities, with or without other signs of motor disturbances, may also disappear gradually over a period of many months Where such improvement is not seen after

erage severity of illness, in the control of complications, and of accuracy of diagnoses in different epidemics make it extremely difficult to arrive at a conclusive figure for the incidence of sequelae, not only in Japanese encephalitis but in other forms as well. For example, the apparent virulence of the 1938 outbreak of Eastern E E, in which only 7 of the 34 diagnosed cases survived (and of these only two made a complete recovery) may be due to the fact that the majority of cases were infants and young children. It is also possible that in this, the first epidemic of its kind, many mild cases were overlooked. On the other hand, in the Manitoba outbreak of Western E E. of 1941 (1), the average severity was comparatively mild,

TABLE 4

Analysis of Findings in Eleven Patients with Neurological Sequelae (Japanese Encephalitis on Okinawa, 1945, Re-examined 10–12 Months After Onset)

PATIENT	AGE	SEX	FINDINGS	MENTAL STATUS
1	4	M	Athetoid gesture, rt arm	Normal
2	5	M	Weakness, lt arm and leg	Mental retardation
3	5	M	Quadriplegia	Advanced deterioration
4	7	F	Rt. hemiplegia, partial paralysis cranial nn. 10, 11, 12	Aphasia
5	9	F	Normal	Aphasia
6	13	M	Incoordination, rt hand	Normal
7	14	M	Weakness, rt arm and leg	Normal
8	14	F	Weakness, lt leg	Mental retardation
9	15	F	Normal	Mild personality change
10	15	F	Normal	Mental retardation
11	21	F	Incoordination, both legs	Mental retardation

From Simpson and Meiklejohn. Am J. Trop Med, 27. 727, 1947

and the simultaneous presence of poliomyelitis undoubtedly led to unavoidable errors in classification, especially in the borderline cases of "aseptic meningitis". These two factors may account for the low incidence of serious residua mentioned by Adamson and Dubo.

It is fair to assume that the incidence of permanent residua among survivors of epidemic summer encephalitis does not, as a rule, exceed 10 to 20 per cent

Post-encephalitic Parkinsonism, so common after von Economo's disease, is an extremely rare sequel to the arthropod-borne encephalitides Its absence is perhaps a more significant differential-diagnostic point, if only in retrospect, than any other clinical criterion

COMPLICATIONS

Complications frequently encountered, especially in severe cases of arthropod-borne encephalitis, are those of any other serious debilitating illness

unattended and is incontinent, unresponsive, and unable to communicate in any way". The contortions, contractures and extreme motor deterioration are well illustrated in the photograph. The condition of this child remained unchanged when re-examined ten months after onset (120).

Such extreme residua are exceptional, but various other symptoms often follow in the wake of summer encephalitis. Among these are headaches, sleep disturbances, nervousness, personality changes, impairment of memory, tremors, spastic pareses, and a variety of focal neurological manifestations. In 1946, Simpson and Meiklejohn re-examined 38 of 77 survivors



FIG 1 FIVE-YEAR-OLD OKINAWA NATIVE, 51 DAYS AFTER ONSET OF JAPANESE ENCEPHALITIS

(Appreciation is expressed to Dr Leon Lewis for permission to reprint this illustration from Lewis, L et al , in Arch Neurol and Psych , 57: 430–463, 1947)

of the 1945 epidemic of Japanese encephalitis on Okinawa and Heanza; 11 of the 38 showed the residua in table 4 The remaining 27 had recovered completely, many of them giving histories of gradual improvement continuing for weeks or months after onset (120).

INCIDENCE OF SEQUELAE

The incidence of sequelae in this Okinawa outbreak was higher than that reported for other outbreaks of Japanese encephalitis by Kaneko and Aoki (64). However, Simpson and Meiklejohn point out that the Okinawa epidemic differed from previous outbreaks by its low mortality rate, and they suggest that many patients with severe damage to the CNS who survived with residua did so because of adequate hospital care but would have succumbed without it. Considerations of this kind variations in av-

erage severity of illness, in the control of complications, and of accuracy of diagnoses in different epidemics make it extremely difficult to arrive at a conclusive figure for the incidence of sequelae, not only in Japanese encephalitis but in other forms as well. For example, the apparent virulence of the 1938 outbreak of Eastern E.E., in which only 7 of the 34 diagnosed cases survived (and of these only two made a complete recovery) may be due to the fact that the majority of cases were infants and young children. It is also possible that in this, the first epidemic of its kind, many mild cases were overlooked. On the other hand, in the Manitoba outbreak of Western E.E of 1941 (1), the average severity was comparatively mild,

TABLE 4

Analysis of Findings in Eleven Patients with Neurological Sequelae (Japanese Encephalitis on Okinawa, 1945, Re-examined 10–12 Months After Onset)

PATIENT	AGE	SEX	FINDINGS	MENTAL STATUS
1	4	M	Athetoid gesture, rt. arm	Normal
2	5	M	Weakness, lt arm and leg	Mental retardation
3	5	M	Quadriplegia	Advanced deterioration
4	7	F	Rt hemiplegia, partial paralysis cranial nn. 10, 11, 12	Aphasia
5	9	F	Normal	Aphasia
6	13	M	Incoordination, rt hand	Normal
7	14	M	Weakness, rt. arm and leg	Normal
8	14	F	Weakness, lt leg	Mental retardation
9	15	F	Normal	Mild personality change
10	15	F	Normal	Mental retardation
11	21	F	Incoordination, both legs	Mental retardation

From Simpson and Meiklejohn: Am J. Trop Med, 27: 727, 1947.

and the simultaneous presence of poliomyelitis undoubtedly led to unavoidable errors in classification, especially in the borderline cases of "aseptic meningitis". These two factors may account for the low incidence of serious residua mentioned by Adamson and Dubo.

It is fair to assume that the incidence of permanent residua among survivors of epidemic summer encephalitis does not, as a rule, exceed 10 to 20 per cent

Post-encephalitic Parkinsonism, so common after von Economo's disease, is an extremely rare sequel to the arthropod-borne encephalitides. Its absence is perhaps a more significant differential-diagnostic point, if only in retrospect, than any other clinical criterion

COMPLICATIONS

Complications frequently encountered, especially in severe cases of arthropod-borne encephalitis, are those of any other serious debilitating illness

unattended and is incontinent, *unresponsive, and unable to communicate* in any way". The contortions, contractures and extreme motor deterioration are well illustrated in the photograph. The condition of this child re*mained unchanged* when re-examined ten months after onset *(120)*

Such extreme residua are exceptional, but various other symptoms often follow in the wake of summer encephalitis. Among these are headaches, sleep disturbances, nervousness, personality changes, impairment of memory, tremors, spastic pareses, and a variety of focal neurological manifestations. In 1946, Simpson and Meiklejohn re-examined 38 of 77 survivors



FIG 1 FIVE-YEAR OLD OKINAWA NATIVE, 51 DAYS AFTER ONSET OF JAPANESE ENCEPHALITIS

(Appreciation is expressed to Dr Leon Lewis for permission to reprint this illustration from Lewis, L et al, in Arch Neurol and Psych, 57: 430–463, 1947)

of the 1945 epidemic of Japanese encephalitis on Okinawa and Heanza; 11 *of the 38 showed the residua in table 4 The remaining* 27 had recovered completely, many of them giving histories of gradual improvement continuing for weeks or months after onset (120).

INCIDENCE OF SEQUELAE

The incidence of sequelae in this Okinawa outbreak was higher than that reported for other *outbreaks of Japanese encephalitis by* Kaneko and Aoki (64) However, Simpson and Meiklejohn point out that the Okinawa epidemic differed from previous outbreaks by its low mortality rate, and they suggest that many patients with severe damage *to the* CNS who survived with residua did so because of adequate hospital care but would [illegible]umbed without it. Considerations of this kind, variations in av-

Blood Hemoglobin and erythrocyte counts are normal. Leukocytosis (10,000 to 25,000 total count) is characteristic of the acute phase. Early in the febrile stage, there is a predominance of neutrophile leukocytes with a shift to the left The count usually returns to normal values at defervescence.

Other clinical laboratory findings are non-contributory in uncomplicated cases. In general, pleocytosis and leukocytosis are the only consistent laboratory findings. Quantitative variations in these two findings are of no prognostic significance, covering an equal range in benign and fatal cases. Gross abnormalities in other clinical laboratory findings (urine, blood, or CSF) point either to another diagnosis or to complications unrelated to encephalitis.

Pathology

GENERAL REMARKS

In uncomplicated cases of arthropod-borne encephalitis, specific lesions due to the action of the infecting viruses are limited to the CNS. Lesions in other organs are non-specific or those of pre-existing disease or newly acquired complications

The variability in the response of nervous tissue to a multitude of noxious agents is necessarily limited. As in many pathological conditions, neuronal and proliferative inflammatory lesions dominate the pathological picture in arthropod-borne encephalitides. Neither basic lesion has any specific characteristic which distinguishes it from that caused by other viral or non-viral agents In particular, the arthropod-borne encephalitides are not characterized by any specific type of cytoplasmic or intranuclear inclusion bodies, in contrast to rabies or herpes-like viruses. This lack of distinguishing marks applies to the entire group Moreover, one cannot differentiate one type of arthropod-borne encephalitis from others. The total number of well-documented post-mortem findings is relatively small and represents a chance selection of a few examples for all forms. Unquestionably, just as in clinical reports so in reports on pathological findings, the issue has been obscured occasionally by the inclusion of unrelated illnesses. The types, intensity, and distribution of lesions may vary because of "non-specific" factors as much as the particular type of virus involved. Some variables, such as age of the patient, duration of the disease, unrelated complicating illnesses, time elapsed between death and autopsy, may be reflected in the predominant or allied pathological findings in different epidemics.

If one makes allowance for variables, one comes to the conclusion that apparent differences in the pathological picture of the various arthropod-borne encephalitides are quantitative at best, and that the basic character

and are of grave significance mainly in elderly patients. They are primarily the effects of inanition and of secondary infections. Bronchopneumonia is common, especially in patients with prolonged coma, and may be the cause of death. Urinary retention or incontinence leads to ascending infection of the urinary tract. Decubitus is common.

Other complications are usually due to the accentuation of pre-existing conditions, such as malnutrition, arteriosclerosis, hypertension, chronic heart disease, chronic nephritis, and infectious diseases or parasitic infestations Such complications are apt not only to aggravate the prognosis, but tend to obscure the diagnosis

Little is known about the effect of infection with these viruses on the course of *pregnancy* in human beings In the St. Louis outbreak of 1933, interruption of pregnancy occurred in three cases, all near term (94). Medovy (79) reported on three infants in whom encephalitis due to Western equine virus appeared three to four days after birth, with simultaneous mild febrile illness in the mothers. This is reminiscent of the recent findings of Burns (11), who isolated Japanese encephalitis virus from the CNS of swine which were either still-born or died in the neonatal period While their mothers seemed to be healthy, the baby pigs showed pathological findings typical of encephalitis. There is, however, no report of congenital malformation acquired by offspring of mothers who had encephalitis during the early stages of pregnancy, akin to those reported for certain exanthematous virus diseases.

Clinical Laboratory Findings

With regard to clinical laboratory findings, no significant difference exists between the various forms of summer encephalitis.

Cerebrospinal Fluid The fluid is usually clear and colorless, but may be slightly turbid. Pellicle formation is rare Pressure may be normal or slightly increased Pleocytosis is one of the most consistent findings in all forms The number of cells varies over a wide range, from a slight increase to 1,000 or more per cmm The majority of cases probably have, at one time or another during the acute stage, 100 to 500 cells per cmm. Pleocytosis may *increase* during the acute phase It may persist for weeks in severe cases, but usually the count returns to normal values by the 20th day after onset The differential count usually shows preponderance of polymorphonuclear leukocytes early in the acute phase with subsequent shift to 80 to 100 per cent mononuclear cells

Protein is either normal or slightly increased Greater increases have been noted later during convalescence or if the disease progresses after the acute febrile stage. Values found for CSF sugar are in the normal range, but a few exceptions in either direction have been noted No other consistent deviations from normal findings have been published.

meshes of the pia-arachnoid. This is seen mainly in cases surviving for more than a week.

Neuronal damage runs the entire range, from mild chromatolytic changes to complete autolysis and disappearance of cells, with all intermediate degrees of neuronal degeneration. Sometimes only a few neurons are involved in a given area, but in other places there may be circumscribed foci in which all the neurons are necrotic. In the latter case, the area is acellular and spongy, the microscopic counterpart of patches of discoloration seen in gross cross sections. Instead of this denuded punched-out appearance, affected areas may show diffuse infiltration by hematogenous elements, both of the neutrophile and the lymphatic type. Neutrophile polymorphonuclear cells may predominate in early cases, later to be replaced largely by lymphocytes and large macrophages. Mononuclear cells with large, irregular nuclei are common and have been variously interpreted as being mobilized Hortega cells or cells derived from the blood stream and from the vascular adventitia (51). This latter type of cell is found not only in necrotic parenchymatous nodules which may have no apparent relationship to blood vessels, but also in perivascular infiltrates throughout the CNS. Perivascular cuffing of variable intensity is common and this may extend into the white matter adjacent to an involved area of gray matter. Neuronal degeneration or destruction may, instead of leading to a simple "outfall" of the cell, be associated with microglial replacement or with outright neuronophagia, the latter mainly by neutrophilic leukocytes or by phagocytic, large mononuclear cells. This type of cellular reaction may reach such a high degree of intensity and the interstitial tissue may be so necrotic, that the lesions resemble miliary abscesses (94, 146) In such lesions, myelin stains reveal total loss of fibrillar structures. Otherwise, demyelination is not a prominent feature. It may be present, however, in white matter next to damaged portions of gray matter.

Necrosis of neurons and interstitial tissue may lead to circumscript foci of encephalomalacia. In older cases, calcium salts may be deposited in these lesions and elsewhere in damaged tissue The eventual fate of lesions is suggested by the findings of Zimmerman in cases surviving for 35 to 52 days after onset of Japanese encephalitis He noted (a) scarring (gliosis) of smaller areas of encephalomalacia or (b) persistence of larger areas of tissue destruction in forms of cysts, after removal of debris by phagocytosis

Distribution of Lesions

Lesions of all types described are found most commonly in the gray matter of the cerebral cortex, basal ganglia, pons, medulla, cerebellum and spinal cord Haymaker and Sabin (51) made a painstaking study of the

of the lesions encountered is the same for all. Therefore, the essential pathological features of the entire group will be summarized, based largely on the detailed descriptions given for each form. (St. Louis—McCordock et al (78); Japanese—Kaneko and Aoki (64), Zimmerman (146), Haymaker and Sabin (51); Eastern equine—Farber et al. (31); Western equine—Baker and Noran (5), Quong (95); Russian spring-summer—Silber and Soloviev (119).)

GROSS PATHOLOGY

The CNS of patients dying in the acute stage of encephalitis shows little on gross inspection. The meninges are clear, there is no visible exudate or hemorrhage. Meningeal and cerebral blood vessels are congested. Diffuse or blotchy pink discoloration of the gray matter of the cortex and of the basal ganglia, pons, medulla, and spinal cord has been reported. This type of discoloration may give the cross section of the spinal cord a gross appearance similar to that seen in poliomyelitis.

In patients dying after prolonged illness, the gray matter of the brain and cord may show far more impressive gross abnormalities. Zimmerman (146), *in his description of three cases of Japanese encephalitis* dying more than one month after onset, pointed to circumscript pale or granulated and gritty patches scattered through the cortical gray matter Sectioning through such lesions gave the sensation of cutting through calcified tissue. Similar, but larger, patches were present in the central gray matter, chiefly the *pallidal nuclei, thalami, red nuclei,* and *substantiae nigrae* These are the structures most heavily involved with microscopic lesions, and in advanced cases they may contain encephalomalacious cysts.

HISTOPATHOLOGY

Types of Lesions

Lesions are primarily limited to gray matter The white matter is probably affected only by extension of lesions in neighboring gray areas All levels of the brain and spinal cord display some effects of the infection These diseases are identifiable as non-suppurative, acute or subacute, polioencephalomyelitides In addition to injury to the neurons, there are diffuse areas or perivascular and nodular parenchymatous foci of inflammatory infiltration

The leptomeninges show varying degrees of diffuse and perivascular infiltrations. Infiltrating cells are predominantly lymphocytes but in very early cases there may be a predominance of polymorphonuclear leukocytes paralleling the shift seen in differential cell counts in cerebrospinal fluid Large mononuclear cells and macrophages may be interspersed in the

(6) Significant clinical laboratory findings in arthropod-borne encephalitis are limited to increased spinal fluid and blood white cell counts Spinal fluid changes, such as persistent presence of erythrocytes or polymorphonuclear cells, or presence of bacteria and persistent changes in spinal fluid chemistry, especially in sugar content, are incompatible with a diagnosis of arthropod-borne encephalitis. So are gross abnormalities in blood, other than leukocytosis, and in urine. Abnormal laboratory findings in an otherwise typical case may, however, be due to complicating or pre-existing conditions.

(7) Pathological findings are those of a diffuse, non-suppurative, non-bacterial meningo-polio-encephalomyelitis with neuronal and infiltrative reaction Extensive involvement of the white matter, either in the form of demyelination or of widespread infiltrative lesions, is not a manifestation of the summer encephalitides

(8) Finally, one might look upon the geographic locale as supporting differential-diagnostic evidence pro or con The occurrence of suspected cases in an area known to be endemic is a particular challenge to one's diagnostic acumen, rather than an easy key to the proper diagnosis because here the temptation to identify everything with the endemic agent is naturally great. Conversely, there is the possibility of known or unknown types of arthropod-borne encephalitides invading previously unaffected areas

The differential-diagnostic criteria just enumerated are admittedly sketchy and incomplete. They are intended as a guide to facilitate the screening of cases While the clinical syndrome of acute encephalitis can be simulated by many heterogeneous illnesses, fortunately most of these can be readily identified or excluded. In this category are some which affect the CNS primarily, such as bacterial meningitis and brain abscess, brain tumors, cerebral hemorrhages, due to accident or vascular disease, and psychoneurotic episodes In acute infectious diseases due to bacteria, viruses, or rickettsiae, or in parasitic diseases with secondary involvement of the CNS, there are, as a rule, signs, symptoms, and laboratory findings referable to other organ systems which are decisive. Chemical or bacterial intoxications of various types may be extremely confusing and are perhaps among the most difficult alternative diagnoses to rule out If the history is of no help, the only early distinguishing finding may be absence of pleocytosis In metabolic and endocrine disorders with acute neurological episodes, the differentiation may depend entirely on clinical laboratory findings

Thus, the relative accuracy with which viral encephalitis is differentiated from a vast array of non-viral neurological diseases depends largely on the quality and thoroughness of clinical and laboratory analysis.

topography of lesions in the CNS of a patient dying about ten days after onset of Japanese encephalitis. They found "nodules" and perivascular infiltration scattered throughout the gray matter. Particularly hard-hit were the thalamus, the substantia nigra, and the basal nuclei. In these structures they noted a striking tendency for the neuronophagic nodules to be confluent. Other areas of gray matter were also heavily and diffusely involved. In the cerebellum, the most notable feature was an almost complete wiping-out of the Purkinje cell layer in some regions.

While the study by Haymaker and Sabin was restricted to a single case, their findings confirm other reports. Theirs is the most carefully examined representative case of arthropod-borne encephalitis of any type.

Differential Diagnosis

GENERAL CONSIDERATIONS

The seasonal encephalitides pose diagnostic problems of the greatest variety. With their clinical severity running the gamut from mild, grippe-like syndromes to fulminating, rapidly fatal forms or to chronic vegetating deterioration—with all intermediate degrees of severity and duration—there is almost no limit to the diversity of conditions with which these maladies have been or could conceivably be confused. An attempt at detailed enumeration of differential-diagnostic problems and challenges would be futile. Instead, recapitulation of a few guiding considerations may be helpful.

(1) Arthropod-borne encephalitis occurs during the late spring and through the summer or early fall, coinciding with the season of peak prevalence of the transmitting arthropods. At other times of the year, even if the clinical picture is suggestive, there are many other possibilities that should be explored first.

(2) Arthropod-borne encephalitides are *acute* diseases. The onset of neurological manifestations in particular is sudden and dramatic and associated with fever.

(3) Neurological signs vary in type and severity, but they are characteristically (a) meningitic manifestations (nuchal rigidity, pleocytosis), (b) if anything else, signs of primary cerebral damage, notably mental disturbances and signs of higher motor neuron involvement (spastic pareses, purposeless movements, convulsions, etc.)

(4) Flaccid paralysis of the extremities, in the absence of primary cerebral signs, is not an expected finding in summer encephalitis.

(5) Spinal fluid pressure is increased but not excessively so, and there is no evidence of ventricle block (negative Queckenstedt). Papilledema, if present, is slight.

Neurological Complications of Other Infectious Diseases; Post-infection and Post-vaccination (-immunization) Encephalitis

Recent or simultaneous infectious illness, notably one of the exanthematous diseases of childhood, or recent smallpox vaccination or other types of immunization constitute the most significant clue The clinical course of these forms of encephalitis is variable. Grouping them together here does not imply that their pathogenesis is related. Indeed, it has already been pointed out that neurological complications of certain virus diseases may sometimes be due to the direct effects of the virus on the CNS, as in mumps. Most cases of post-infection and post-immunization encephalitis, however, appear to be due to an agent or agents not related to the known virus encephalitides This is suggested by the fact that they are leuko-encephalitides, characterized by widespread demyelination and by infiltrative lesions in the white matter, and by the fact that virus isolation from CNS tissue with a predominance of such lesions has failed invariably.

Infectious Polyneuritis (Guillain-Barré Syndrome)

This is characterized by paresthesias and pain, loss of deep reflexes and of some sensory perception, and by paralysis, usually of the ascending (Landry) type. Paralysis may be restricted to certain cord segments, such as the shoulder girdle—reminiscent of Russian spring-summer encephalitis Fever is slight or absent The cerebrospinal fluid shows increased protein but a normal cell count. The disease may be of prolonged duration, three to four months, but the prognosis is relatively favorable There is no evidence of cerebral damage. The etiology is unknown, and repeated efforts have failed to yield a virus.

Other Viral Encephalitides and "Aseptic Meningitis"

No reliable criteria can be offered for the clinico-pathological differentiation of arthropod-borne from other forms of viral encephalitis. This is true not only for sporadic cases but also in the framework of a known epidemic The positive identification on clinical grounds alone is beyond reach especially in cases of "aseptic meningitis", a syndrome which can be caused by many heterogeneous agents. The task of screening this type of material for specific viruses falls entirely to the laboratory equipped to carry out virus isolations or serological tests. A later section will give brief descriptions of these procedures, with special emphasis on an evaluation of their scope and limitations under various conditions.

SPECIAL DIFFERENTIAL DIAGNOSTIC PROBLEMS

The points of distinction are finer when it comes to differentiation of viral encephalitis from other virus infections of the CNS or from certain neurological syndromes of unknown etiology. Of these, the following merit somewhat more detailed discussion:

Poliomyelitis

The seasonal incidence coincides with that of arthropod-borne encephalitides, and several instances of concurrent epidemics of the two types of infection have been reported Thus, poliomyelitis and Japanese encephalitis occurred simultaneously among American troops in the Far East, and the large outbreak caused by Western equine encephalitis virus in 1941 in Manitoba was complicated by a simultaneous epidemic of poliomyelitis (26). In borderline cases of "aseptic meningitis", differentiation is almost impossible without recourse to specific diagnostic tests. In more typical cases, the difference is reasonably characteristic. In poliomyelitis, cerebral symptoms of the type seen in encephalitis are not a presenting feature, even though spasticity in early stages may be associated with lesions in the motor cortex and brain stem (9). More usual in poliomyelitis are *late* cerebral symptoms due to anoxia in cases with bulbar involvement. Progressive flaccid paralysis, especially in the absence of primary cerebral impairment, points to poliomyelitis, but it occurs sometimes in the course of viral encephalitis, especially in the Russian spring-summer variety. Histologically, poliomyelitis and primary viral encephalitis are similar The lesions in the spinal cord are indistinguishable, but involvement of cerebral cortex, cerebellum, and basal ganglia in poliomyelitis is much less pronounced than in encephalitis

Von Economo's Encephalitis

Epidemics of this form usually occurred in winter and early spring, and this difference in seasonal incidence is the most immediate distinguishing mark. The onset of von Economo's disease is often more gradual High fever and pronounced meningeal signs are not invariable as in arthropod-borne encephalitis. Ophthalmoplegia and sequelae in form of Parkinsonism, both very frequent in the Economo type, are conspicuous by their absence in summer encephalitis The lesions found in the CNS in the two forms are very similar and differ perhaps only quantitatively, being somewhat more discrete in the von Economo type. The differentiation of the latter from the "newer" forms of epidemic encephalitis has to be based on specific diagnostic laboratory tests, especially in sporadic cases.

contact from man to man, are particularly prevalent in densely populated communities Not so the summer encephalitis viruses. The main foci of endemicity are rural areas which abound in various susceptible animal species and in arthropods capable of transmitting the viruses and harboring

TABLE 5

Distribution of Known Arthropod-borne Encephalitides by Continents

TYPE	NORTH AMERICA	CENTRAL AMERICA	SOUTH AMERICA	OCEANIA AND AUSTRALIA	ASIA	EUROPE
St Louis	Central US Western US Southwest					
Western EE	Central and Western US and Canada		Argentina?			
Eastern EE	Central and Eastern US and Canada	Mexico Cuba Panama	Brazil			
Venez EE		British West Indies	Venezuela			
Japanese				Australia Philippine Isl. Guam	Japan Ryukus Formosa Eastern China Manchuria Maritime USSR Korea	
Russian spring-summer					Far East USSR Siberia	European USSR?
Louping Ill						European USSR? Central Europe British Isles

them for long periods of time Close symbiosis of man with domestic animals seems to create favorable epidemiological background Hence, human epidemics are most apt to occur in agricultural regions, in mosquito-infested valleys or swamp lands, or, in the case of tick-transmitted Russian spring-summer encephalitis, in densely wooded territory.

The complexity of the ideal requirements for preservation and activity of each virus—i.e., the need for a susceptible animal reservoir and vectors—is probably the chief factor limiting its dissemination Undoubtedly, in-

Epidemiology

GEOGRAPHIC DISTRIBUTION

Much knowledge of the epidemiology of the human arthropod-borne encephalitides, especially their geographic distribution, is not based on study of the disease as it occurs in man In many areas in which one or the other of the causative viruses is known to exist, human cases have never been seen or have not occurred in epidemic proportions. Yet, their wide dissemination has been established either by recognition of illness they cause in animals or by serological surveys of the animal and human populations in enzootic or endemic regions. Almost every large area which has been subjected to a thorough search during the past few years has yielded evidence for the presence of one or several viruses belonging, or suspected of belonging, to the arthropod-borne encephalitis family. With an ever-increasing degree of attention paid to serological surveys, certain geographic boundaries have turned out to be less sharp than they were previously thought to be.

If one considers only the viruses of proved ability to cause human encephalitis, their distribution suggests restriction to continental confines (table 5). Thus, neither St. Louis nor any of the equine encephalitis viruses has ever been found outside of the Western hemisphere. Japanese encephalitis virus, endemic in the countries bordering on the China Sea and Japanese waters, has spread as far as Australia, the Philippine Islands, and Guam Russian spring-summer encephalitis virus and its close relative, louping-ill virus, are disseminated contiguously through the Eurasian continent. Certain parts of Africa, not known to harbor any of the proved seasonal encephalitis viruses, have yielded a multitude of agents which may belong in that group (see above, table 2) One of these, West Nile virus, is immunologically related to St Louis and Japanese viruses.

The co-existence of two or more viruses in the same region is not restricted to immunologically related combinations. At least five states of the United States harbor both Eastern and Western equine viruses, namely, Alabama, Arkansas, Florida, Michigan and Texas. Evidence for the additional presence of St. Louis encephalitis in Texas has been reported (41). Both St Louis and Western equine types are endemic not only in the same states but in the same localities, e g , the Yakima Valley in Washington (40) and Kern County, California (12)

There must be common denominators which stamp certain select localities as particularly well suited for the continued preservation and propagation of these viruses If man were their chief carrier and transmitter, one would expect the same world-wide distribution that has been attained by mumps, measles, influenza, and many other viruses. These, spreading by

tion of monkeys with a mixture of Western equine and St. Louis viruses led to dual antibody production in the serum, although it appeared that the CNS was reached and affected only by the equine virus (see below under experimental infection and immunity).

SEASONAL INCIDENCE

The seasonal incidence of each form of arthropod-borne encephalitis is sharply defined. Since the epidemic curves closely parallel the rise and fall in suspected vector population, the timing of each outbreak shifts according to the climatic conditions existing in the particular locality. Thus, epidemics of Japanese encephalitis in the Japanese chain of islands generally move northward, beginning about June in the southern regions and extending to the northern parts of Honshu about late August to September

The months of peak prevalence have been reported for the various forms as follows (in man only):

St Louis encephalitis	late August to early Sept.
Japanese encephalitis	late June to September
Western equine encephalitis	mid-July to September
Eastern equine encephalitis	mid-July to September
Russian spring-summer enc	mid-May to mid-July

INCIDENCE ACCORDING TO AGE, SEX, OCCUPATION

In discussing the incidence of arthropod-borne encephalitides, one has to distinguish between two phases, (a) morbidity rate and (b) infection rate Serological evidence indicates that in endemic regions, large proportions of the population have passed through nonapparent infections with the entrenched virus The incidence and geographic distribution of neutralizing antibody in the "normal" population is generally considered to be a sensitive measure of the past prevalence of the virus. Assuming this to be a reliable criterion, and assuming also an equal ability of people to respond to infection with antibody formation regardless of age, one must postulate that variations in incidence of antibody among different segments of the population reflect variations in intensity or frequency of past exposure Assuming, again, that presence of neutralizing antibody acquired through natural infection has any meaning in terms of the individual's response to re-infection, one would expect the morbidity rate to be highest in those age groups or other segments which have had the least past exposure to the virus.

If all these assumptions are correct, one should expect considerable variations in the epidemiological pattern from one outbreak to another Conclusive evidence along such lines is still scanty for most of the arthropod-borne encephalitides, but an increasing amount of information, especially on the Japanese form, bears out some of these postulates

crease in the amount and speed of travel and transportation, even to and from areas previously fairly isolated from contact with the outside world, will increase the danger of spread of these viruses into suitable virgin territory. Thus, in 1948, Japanese encephalitis suddenly appeared on Guam, "almost 2,000 miles further east than it had been suspected to extend previously" (44). Hammon also cites the examples of two Americans who apparently became infected with Japanese virus shortly before sailing from Okinawa and Yokohama, respectively. Both developed the disease on shipboard, the second case just one day before arrival in San Francisco, where he died a few days later. The implications of these isolated instances are obvious.

Fortunately, man appears to be a relatively unimportant and accidental link in the complex ecology of these viruses. If, as appears almost certain, the seasonal encephalitides are indeed conveyed mainly or exclusively by the bites of arthropods, then the possibility of intercontinental transportation of infected vectors would entail far greater danger of dissemination than human travel.

Of special interest in this connection is a comparison between the geographical distribution of two other arthropod-borne virus diseases of man, namely, yellow fever and dengue, with that of the arthropod-borne encephalitides. This is especially appropriate in view of the many similarities between these two groups of viruses which have been found most recently to include an immunologic relationship (104). The natural host range of yellow fever and dengue appears to be limited to monkeys, human beings, and mosquito vectors. Hence, the conditions required for their survival and activity are relatively simple to fulfill and this may explain why both diseases have at various historical periods been endemic or epidemic in many different parts of the world As far as is known, all strains of yellow fever virus are immunologically closely related, if not identical, regardless of their place of origin. Dengue, on the other hand, is caused by at least two immunologically distinct though related types of virus, both of which may co-exist in the same region (e g , New Guinea in 1943) (110). Despite the wide dissemination of dengue and yellow fever viruses and their similarities, their *simultaneous endemic* presence in the same region has never been established. The suggestion has been made that the two viruses may be mutually exclusive since it has been found experimentally in man, monkeys, and mosquitoes that infection with one may interfere with the successful establishment of the other (102) In view of the fact that interference between certain of the arthropod-borne encephalitic viruses can also be shown experimentally (27, 57, 117, 118), it may not be amiss to speculate on the possibility of a similar mechanism contributing to the localization of certain encephalitis viruses Indeed, Howitt (56) found that infec-

anese encephalitis, especially those on Okinawa studied by American observers, the highest morbidity rates in natives were found among the young, especially in the 0 to 13-year age group (136). In 1948, approximately 8,000 cases of encephalitis were reported in Japan. The highest incidence, 117 per 100,000, was found in the 6 to 10-year age group, while

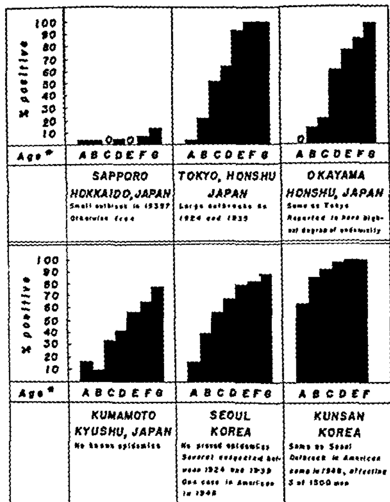


FIG 2 AGE DISTRIBUTION OF NEUTRALIZING ANTIBODY AGAINST JAPANESE ENCEPHALITIS VIRUS IN "NORMAL" HUMAN BEINGS LIVING IN DIFFERENT PARTS OF JAPAN AND KOREA, WITH DATA ON THE HISTORY OF EPIDEMICS UP TO NOVEMBER AND DECEMBER 1946, WHEN THE SPECIMENS WERE COLLECTED

* Age groups (according to Western custom) A, 0–3 years, B, 4–8 years, C, 9–13 years, D, 14–18 years, E, 19–38 years, F, 39–58 years, G, 59 years and over

(Data and modified charts from Bawell et al , Am J Hyg , 51: 1-12, 1950, Deuel et al , Am J Hyg , 51: 13–20, 1950)

Age Incidence

Japanese Encephalitis in Endemic Regions: Epidemiological and diagnostic surveys have been conducted since 1945 by Sabin and his associates and by others in various sections of Japan, Okinawa, Korea and China. Particular stress was placed on attempts to correlate variation in the recorded past incidence of human encephalitis with the antibody pattern as disclosed by neutralization tests The diagrams in figure 2 have been modified from papers by Bawell et al. (6) and Deuel et al. (22). From these data, obtained in 1946, the following points emerge: (a) Only in one area, Hokkaido, was the absence of recognized clinical cases reflected by negligible incidence of antibody (b) In Kumamoto, the other encephalitis-"free" region, there was a suggestion of continued dissemination of the virus over many years resulting in gradual, cumulative acquisition of antibody upon prolonged residence in an area of obviously low-grade endemicity. (c) Absence of large reported epidemics in Japan since 1939 was reflected by a very low incidence of antibody among infants and children up to eight years of age in the Tokyo area. Here, the incidence was close to 100 per cent among those who had lived through all the large epidemics reported since 1924, i.e., those 20 years and older. (d) The findings in the Kunsan district of Korea were most revealing and significant in that over 60 per cent of the samples obtained from infants in the 0 to 3-year age group and 85 per cent of the 4 to 8-year group were positive This is the only one of the areas studied in which cases of Japanese encephalitis were definitely identified in 1946 (111). Similarly, the pattern on Okinawa underwent an important change between the time of the outbreak of August 1945 and December 1946, as shown in table 6 (22).

These last two items justify the conclusion that occurrence of any clinically recognizable cases in a given area—small though their number may be—reflects an experience in which a large proportion of the total population may have participated Despite the evidence of wide dissemination of the virus in the Kunsan area, no native cases of encephalitis came to the attention of the investigators It was the occurrence of encephalitis among Americans in that area which suggested that the virus was present and could infect human beings

Actually, simultaneous studies on a variety of domestic animals in these areas revealed that certain species acquired antibody even at times when there was no evidence—direct or indirect—of human infection. Thus the incidence of antibody among human beings is not so much a true index of the prevalence or absence of the virus from the region as an index of its ability to infect or to be conveyed to man

The age incidence of encephalitis must be considered in the light of these basic epidemiological findings In the more recent epidemics of Jap-

RUSSIAN SPRING-SUMMER ENCEPHALITIS. The data on age incidence of this form are necessarily influenced by the specialized conditions under which people are exposed to infection. The incidence is highest (80 per cent of all cases) in the 20 to 30-year group, but this is due to the fact that this group constitutes the main force of forest workers in immediate contact with the transmitting wood tick, *Ixodes persulcatus* (119).

Sex and Occupational Incidence

As in the Russian spring-summer form, so in the other types of arthropod-borne encephalitis the highest incidence may be found among those who are most exposed to transmitting insects. These are usually men in the 20 to 40-year groups, who work the fields, and hence in this range the incidence among men may be higher than among women There is no evidence of any influence of sex on susceptibility.

ANIMAL RESERVOIRS

The three so-called equine encephalitis viruses were first isolated from horses and only later found to be pathogenic for man. This is not surprising in view of the staggering losses which the equine population of the United States and of South American countries suffered prior to the first recognized outbreak in human beings Thus, in 1938, the combined incidence for the Eastern and Western type for the United States was 184,662 in a total population of about 15 million horses and mules The incidence has dropped considerably since the initiation of energetic vaccination programs (82) Where human cases have been recognized, they were preceded by cases of encephalitis among horses Moreover, it has recently been demonstrated (10) that the Japanese varieties of equine and of human encephalitis are caused by the same virus Burns has shown also that Japanese encephalitis virus is responsible for stillbirth of swine in Japan, although apparently nonpathogenic for adult pigs Finally, louping-ill was primarily known as a disease of sheep rather than man The natural host range of the arthropod-borne encephalitis viruses does not, however, end with these few susceptible species It has become increasingly evident that each of the viruses is capable of infecting many mammals and birds which, through their vast number alone, may play a far more important role as links in the infection cycle than either man or horse.

It is now generally assumed that the viruses in this group are transmitted to man through the bites of infected mosquitoes or, in the case of Russian spring-summer encephalitis or louping-ill, ticks (*vide infra* for a summary of the evidence for their role). It is reasonable, then, to expect that the chief reservoirs of the viruses would be those species which are preferentially bitten by the vectors and in which, at the same time, the viruses

in the 31 to 70-year age group it averaged only 9.1 per 100,000 (144). This is in marked contrast to the increased incidence with increasing age reported for the years 1924 to 1933 in Japan (77).

Such a shift in age distribution might be due to the progressive immunization of the older age groups through nonapparent infection Reports on the age incidence of Japanese encephalitis and of other forms should therefore not be interpreted as inherent characteristics of each type, but rather as reflecting the particular situations in which they were obtained

St. Louis Encephalitis. The 1933 outbreak in St. Louis was characterized by progressively higher morbidity rates in each decade of life The incidence in young children was reported as very low (94)

Western Equine Encephalitis The figures for the 1939–1941 out-

TABLE 6

Status of Nonapparent Infection with Virus of Japanese B Encephalitis on Okinawa in December 1946, as Compared with That Found During Epidemic of 1945

AGE GROUP	INCIDENCE OF NEUTRALIZING ANTIBODY			
	December 1946		August 1945 (101)	
	No tested	*Per cent positive*	*No tested*	*Per cent positive*
years				
1–4	37	27	9	0
5–9	42	57	7	0
10–14	41	78	11	55
15–19	32	97		
20 and over	80	91	30	90

Data from Deuel, Bawell, Matumoto and Sabin Am J Hyg, 15: 13–20, 1950

breaks in Kern County, California (12), and for the 1941 epidemics in North Dakota (70) and in Manitoba (26) are in substantial agreement The incidence was high among infants less than one year of age (11, 5 and 8 per cent, respectively, of all reported cases) For the 1 to 19-year groups, the rates were conspicuously lower in Manitoba and North Dakota, but not in Kern County The situation in Yakima Valley, Washington, as reported by Hammon (40) was somewhat different in that the incidence in small outbreaks increased with advancing age In Yakima Valley and in Kern County, serological surveys of the "normal" population were conducted by Hammon and by Howitt The frequency of nonapparent infection was not nearly as high as that found in regions in which Japanese encephalitis is endemic

Eastern Equine Encephalitis The only significant outbreak was characterized by an exceedingly high proportion of cases among small children, especially infants under one year of age (37)

tribution of neutralizing antibody among domestic animals in the Tokyo and Okayama regions of Honshu, Japan. While the numbers of specimens were relatively small, the findings in horse sera are nevertheless striking. Six of seven horses which had lived in the Tokyo area for two years or more had antibody, but none of 20 comparable animals from Okayama did. On the other hand, the incidence of nonapparent infection among the other species, combined with the data obtained simultaneously on human sera (see above, figure 2) indicates that the virus was about equally widely

TABLE 7

Incidence of Neutralizing Antibody Against Japanese Encephalitis Virus Among "Normal" Domestic Animals in Tokyo Area and Okayama Prefecture, Honshu, Japan, October 1946

SPECIES	SUMMERS IN AREA	TOKYO AREA			OKAYAMA PREFECTURE	
		No tested	Per cent positive		No tested	Per cent positive
Horses	1	15	53	64	—	—
	2 or more	7	86		20	0
Cows	1	14	7	12	—	—
	2 or more	2	50		20	45
Goats	1	10	10	10	—	—
	2 or more	10	10		16	50
Pigs	1	16	25	35	—	—
	2 or more	4	75		—	—
Chickens	1	9	0	0	15	0
	2	9	0		—	—

Data from Bawell, Deuel, Matumoto and Sabin Am J Hyg, 51: 1–12, 1950

disseminated in the two areas. Such a difference *in host range of the agent* in two areas suggests (a) different transmitting arthropods with *different* biting habits, (b) *quantitative differences in the degree or frequency of* exposure, (c) a mode of transmission other than biting arthropods, (d) variations of the viral strains Whether or not any of these domestic species at any time constitutes the *main* reservoir of virus still remains to be determined.

In the case of Russian spring-summer encephalitis, it has been shown that a large number of wild rodents and birds are infected in nature without showing ill effects (119) It is likely that the wood tick, *Ixodes persulcatus*, picks up the virus from woodland animals and birds, but horses and cows have also been found to possess antibodies

persist in the peripheral blood in sufficiently high concentration to infect other arthropods.

This condition is fulfilled, in the case of the equine and St. Louis encephalitis virus, chiefly by wild and domestic birds. From their blood, these viruses can be isolated for several days and chicken-to-chicken transmission of the viruses by *Culex* mosquitoes has been amply demonstrated in laboratory tests (46, 47). This confirms suspicion held on epidemiological grounds. It had been found previously that a high percentage of wild and domestic birds and fowl in endemic areas possessed neutralizing antibodies to the Eastern, Western, or St. Louis viruses (45), indicating that they had passed through nonapparent infections with these viruses. The incidence of antibody among a variety of wild or domestic mammals other than horses and mules appears to be somewhat lower Hammon (42) has critically reviewed the evidence favoring the contention that birds and fowl constitute the main vertebrate reservoirs for the American encephalitis viruses, but has also mentioned certain instances in which it was not possible to incriminate avian species. Indications are that the ecological background of epidemics can vary a great deal according to the locality involved and its fauna One would expect such an adaptability in the case of viruses of such an exceptionally wide host range.

In this respect, recent studies on the incidence of naturally acquired nonapparent infection of various wild and domestic species with Japanese encephalitis virus have been particularly revealing Avian species appear to play a negligible role in the natural infectious cycle of this agent (6, 22, 101, 136), even though chickens are known to be infectible and able to transmit the virus to biting mosquitoes (48). This apparent contradiction may be resolved by considering that the mosquito believed to be the chief vector of Japanese encephalitis, *Culex tritaeniorrhynchus*, feeds chiefly on certain mammalian domestic animals and only rarely on chickens (112). In contrast, *Culex tarsalis*, believed to be the chief vector of Western equine and St. Louis encephalitis viruses, feeds preferentially on fowl (98). In areas in which the Japanese virus is endemic, it has been a consistent finding that a high percentage of domestic mammalian species but not fowl had antibody. The most significant appear to be horses, cattle, pigs, and goats—all species living in close contact with the native human population (6, 109, 136). Sabin and his collaborators showed that the number of years spent by the horses in endemic regions had a marked influence on the incidence of antibody among them. Variations in the enzootic behavior of the virus from one locale to another are suggested by the fact that in two areas in which the antibody patterns of the human populations were closely analogous, those of the domestic animals were found quite different. This is illustrated by the data presented in table 7 which indicate the dis-

tribution of neutralizing antibody among domestic animals in the Tokyo and Okayama regions of Honshu, Japan. While the numbers of specimens were relatively small, the findings in horse sera are nevertheless striking Six of seven horses which had lived in the Tokyo area for two years or more had antibody, but none of 20 comparable animals from Okayama did On the other hand, the incidence of nonapparent infection among the other species, combined with the data obtained simultaneously on human sera (see above, figure 2) indicates that the virus was about equally widely

TABLE 7

Incidence of Neutralizing Antibody Against Japanese Encephalitis Virus Among "Normal" Domestic Animals in Tokyo Area and Okayama Prefecture, Honshu, Japan, October 1946

SPECIES	SUMMERS IN AREA	TOKYO AREA			OKAYAMA PREFECTURE	
		No tested	Per cent positive		No tested	Per cent positive
Horses	1	15	53	64	—	—
	2 or more	7	86		20	0
Cows	1	14	7	12	—	—
	2 or more	2	50		20	45
Goats	1	10	10	10	—	—
	2 or more	10	10		16	50
Pigs	1	16	25	35	—	—
	2 or more	4	75		—	—
Chickens	1	9	0	0	15	0
	2	9	0		—	—

Data from Bawell, Deuel, Matumoto and Sabin Am J Hyg, 51: 1-12, 1950

disseminated in the two areas Such a difference in host range of the agent in two areas suggests (a) different transmitting arthropods with different biting habits, (b) quantitative differences in the degree or frequency of exposure, (c) a mode of transmission other than biting arthropods, (d) variations of the viral strains Whether or not any of these domestic species at any time constitutes the *main* reservoir of virus still remains to be determined.

In the case of Russian spring-summer encephalitis, it has been shown that a large number of wild rodents and birds are infected in nature without showing ill effects (119). It is likely that the wood tick, *Ixodes persulcatus*, picks up the virus from woodland animals and birds, but horses and cows have also been found to possess antibodies

From the point of view of human welfare, the most important contribution of the accumulated data is the revelation that man is merely an accidental link in the infectious cycle of the viruses. Indeed, the evidence obtained for Japanese encephalitis suggests that in endemic regions the virus may be widely disseminated in the animal kingdom without infecting man to any significant degree (6, 22). This phenomenon may be related to some of the variables mentioned above, i.e., differences in intensity or nature of exposure, variations in the virus, or also local variations in human susceptibility.

EVIDENCE FOR TRANSMISSION BY BLOOD-SUCKING ARTHROPODS

The following considerations strengthen the argument for arthropod transmission of seasonal encephalitis viruses:

(1) The seasonal curves of epidemics and epizootics parallel the relative prevalence of certain arthropods, usually lagging behind by about two weeks.

(2) Arthropod infestation in endemic or epidemic areas is high and such areas always abound in a variety of warm-blooded host species

(3) Evidence of infection (apparent or nonapparent) is highest in those species which are preferentially bitten by the incriminated arthropods.

(4) The epidemic character of the disease—preferential involvement of agricultural or woodland regions and of people working in the field or forest, infrequency of familial contact infection—can best be explained on the basis of arthropod transmission

(5) The demonstration in the laboratory that naturally or experimentally infected arthropods can infect laboratory, wild or domestic animals

(6) The repeated recovery of virus from trapped arthropods.

In addition to these, there are more theoretical or teleological considerations which make it likely that some common factor connects the variety of species which are hosts of the viruses. It is difficult to think of a common denominator other than biting insects

Direct evidence, i e , isolation of the viruses from arthropods caught in endemic or epidemic areas, has led to incrimination of the arthropods listed in table 8 as actual or potential transmitters of the diseases. In addition, many other species are capable of transmitting one or the other of the viruses under experimental conditions So much work has been published on this phase that a detailed account would be impossible within the limits of this paper. Reviews of the subject have been published by Hammon (42) and by Reeves (97) and should be consulted for more complete information dealing with this question

Suffice it to point out that arthropods, once infected, remain infectious for the remainder of their lives. They acquire the ability to transmit the

TABLE 8

Arthropod Species from Which Encephalitis Viruses Have Been Isolated (Natural Infection of Wild-caught Batches)

VIRUS	MOSQUITOES	BIRD OR FOWL MITES	CHICKEN LICE	BLOOD-SUCKING BUGS	TICKS
St Louis	**Culex tarsalis** Culex pipiens Linn Aedes dorsalis	Dermanyssus gallinae Liponyssus sylviarum			
Western equine	**Culex tarsalis** Culex pipiens Linn Culex stigmatosa Culeseta inornata Aedes dorsalis An maculipennis	Derm gallinae Derm americanus Liponyssus sylviarum Lipon bursa		Triatoma sanguisuga	
Eastern equine	Mansonia perturbans	Derm gallinae	Eomenacanthus stramineus		
Venez equine	Mansonia titillans				
Japanese (Russian autumn)	**Culex tritaenorrhynchus** Culex pipiens var pallens An hyrcanus sinensis				
Russian spring-summer					**Ixodes persulcatus**
Louping-ill					**Ixodes ricinus**

Boldface type: Arthropods believed to be chief vectors feeding on man

virus a few days (10 to 14) after engorgement with infected blood, and this *latent period is called the "extrinsic incubation period"*.

The mere knowledge that certain arthropods can carry these viruses and that they are capable—at least under experimental conditions—of infecting vertebrate hosts does not provide a key to another equally important question: Where does the virus come from each year, and how does it maintain itself from season to season?

The first clues to this problem were obtained for louping-ill and Russian spring-summer virus. *Transovarian transmission of these agents by the* tick vectors to their progeny was demonstrated by Russian workers (119), and it was also shown that *Ixodes* could become infected in the larval, nymph and adult stage. In addition virus-infected ticks could hibernate at low temperatures and emerge fully infectious in the spring (131). Thus, the cycle is closed for these viruses by the assumption of a simple tick-vertebrate-tick-vertebrate chain, with man as an "accidental" branch.

The chain of the mosquito-borne encephalitis viruses appears to be more complex. Here again, it may be helpful to point to the contrast between yellow fever and dengue on one hand, and the encephalitis group on the other. The former are endemic in tropical areas which permit adult mosquitoes to hibernate. Most of the areas in which the encephalitides are endemic or enzootic are sub-tropical or moderate, and hibernation of adult mosquitoes is uncommon On the other hand, transovarian passage of virus from mosquito to progeny has not been demonstrated Since all known vertebrate hosts are short-term carriers, the virus being replaced in blood by antibody, one cannot assume that they insure the inter-seasonal maintenance of virus.

This missing link in the infectious cycle of these viruses appears to be provided in the cases of Western, Eastern equine and St. Louis encephalitis viruses by mites which feed mainly on domestic fowl and on wild birds (Genera *Dermanyssus* and *Liponyssus*) In *Dermanyssus gallinae*, *transovarian transmission* of St Louis encephalitis virus has been demonstrated by Smith et al. (122–124), who also showed that the amount of virus circulating in the blood of chickens after bite by naturally infected mites was sufficient to render normal mites or mosquitoes infective for normal chickens.

Blattner and Heys (8) had shown that the dog tick, *Dermacentor variabilis*, could be infected with St. Louis virus and could transmit it to mice by bite. They showed transovarian passage and hibernation of the virus in this arachnid. Smith and her co-workers suggest the existence of two infectious cycles, one in which the maintenance of the virus is assured by a mite-bird-mite cycle with hibernation in mites, and another in which mosquitoes transmit the virus from birds to various other vertebrates, including man.

Similar cycles may be operative in the epidemiology of the equine viruses. Indeed, Syverton and Berry (133) found that the wood tick, *Dermacentor andersoni Stiles*, under experimental conditions could serve as reservoir and transmitter of the Western strain in a manner analogous to that described for *D. variabilis* and St. Louis encephalitis virus. No reliable clue has as yet been obtained concerning the inter-epidemic or -epizootic fate of Japanese encephalitis virus.

Pathogenesis and Immunity

EXPERIMENTAL INFECTION AND IMMUNITY

Many of the mechanisms operating in the pathogenesis of human arthropod-borne encephalitis can be visualized if one understands what happens in infected laboratory animals For the purpose of analogy, the most useful experiments are those in which the supposed natural mode of transmission and infection is simulated as closely as possible. If naturally infected mosquitoes or ectoparasites are allowed to feed on susceptible vertebrates, the amounts of virus subsequently found in the host's circulating blood are very small (124) This indicates that the infecting dose under natural conditions is probably of a low order of magnitude. It stands to reason that the chances of encephalitis developing under such conditions would depend on (a) the ease with which virus can reach the CNS from a peripheral site of entry and (b) the capacity of the virus to multiply and to cause damage in that organ These factors vary for different viruses and for different hosts

Influence of Different Types and Variants of Infecting Viruses

A virus strain can undergo changes as a result of continued passage through laboratory animals and probably as a result of variations in its particular habitat in nature. This explains why different strains of the same type of virus and passage-induced variants may have widely differing degrees of pathogenicity for a variety of hosts infected through various portals of entry. It was shown by Schlesinger (113) that continued brain-to-brain passage of Western equine virus in mice resulted in marked acceleration of its rate of multiplication in mouse brain and consequently in a much more rapid course of the disease after intracerebral inoculation. This was also accompanied by a 100-fold increase in lethal titer of the virus. On the other hand, the relative ease with which a strain can produce encephalitis in experimental animals after extraneural (e.g., subcutaneous, intramuscular or intraperitoneal) inoculation may be entirely unrelated to its encephalitogenic potency after intracerebral inoculation. Thus, certain strains of Western or Eastern equine encephalomyelitis in very low concentrations (10^{-8} or 10^{-9} diluted extract of infected mouse brain) consistently produce fatal illness in adult Swiss mice when introduced directly into the

brain, and yet very large doses are almost entirely innocuous when injected peripherally. If one uses baby mice instead, the minimal infectious dose by the two routes is closely analogous. On the other hand, Russian spring-summer virus is about equally pathogenic for adult mice regardless of the route of inoculation used. Other summer encephalitis viruses occupy a position somewhere between the extremes of laboratory-adapted equine strains and the Russian virus. Variations of this sort need have no bearing at all on the behavior of the viruses in hosts other than the mouse.

Host Factors Affecting the Response to Infection

There are two lines of approach which have thrown some light on this question. These are concerned with a) genetic constitution, b) specific immune response. In addition, a number of hypotheses have been put forward in attempts to explain why in experimental animals as well as in some natural hosts the incidence of encephalitis is so infinitesimal in comparison with that of nonapparent infection.

GENETIC FACTORS Certain species, while supporting viral multiplication outside of the CNS (sustained viremia, followed by antibody formation) escape pathogenic effects of the virus even upon intracerebral inoculation This appears to be true for fowl and other birds infected with St. Louis or the equine viruses, and for certain rodents and other wild or domestic mammals infected with any arthropod-borne encephalitis virus. Hence, *resistance or susceptibility to encephalitis*, as distinct from *susceptibility to infection*, appear to be genetically controlled species traits. The idea that cerebral resistance or susceptibility may be under the control of a single gene has found some experimental support in the work of Webster and Clow (141) and of Sabin (Proc Nat Ac Sc **38**, 540–546, 1952). These investigations revealed that strains of Swiss albino mice can give rise to pure lines which are either resistant or susceptible to St Louis or to Japanese encephalitis or to other viruses and that the results of cross-breeding are in agreement with Mendelian laws governing single gene characters

SPECIFIC IMMUNE RESPONSE Investigation of the rapidity and efficacy of antibody production in response to peripherally injected equine encephalitis virus has furnished a possible rational explanation for the difference in susceptibility between adult and baby mice Morgan (83) found an inverse relationship between susceptibility of mice of different ages to Eastern equine encephalitis virus and their ability to respond to infection with antibody production. She concluded that older mice were "resistant" because the rapid and profuse output of antibody effectively checked virus multiplication before irreversible damage had been done in the CNS, while baby mice succumbed because their immune response was less adequate Schlesinger et al. (116) followed through on this line of reasoning in work

on young and adult rabbits. Here it could be shown that the initial phases of infection were comparable in the young and old, and characterized by viremia and fever (fig. 3). In the young, fatal encephalitis ensues with death occuring before the serum antibody reaches an arbitrarily chosen titer of 1:300 (5th day in fig. 3). By that time, the adult animal has already attained an antibody titer of 1:300 and defervescence occurs before any encephalitic signs are noticeable When such "resistant" adult animals were sacrificed at the height of fever, virus could be demonstrated in their brains—a model for similar findings in human beings dying early in the course of encephalitis and whose serum contains antibody. Later work by Schlesinger (114) suggested that the question of survival or death might be decided by the outcome of a delicate competition, in the CNS, between rate of viral multiplication and the rate of local antibody production In this connection, it is of interest that Howitt (56) demonstrated neutralizing antibody in the spinal fluid of patients convalescent from St. Louis or Western equine encephalitis. In the light of what we know about the "physiological" distribution of antibody between blood and spinal fluid (about 300:1) (35, 84), one may conclude that demonstration of significant amounts in the spinal fluid reflect a specific local response to viral activity within the CNS (63, 114) At any rate, there can be no doubt that the immune response stimulated by the infecting virus outside or within the CNS can be an effective force in checking the progress of the infection, and that species, age or other individual factors can influence this response.

Other Host Factors Differences in the response of a variety of species or of hosts of different ages to encephalitic virus administered by different routes have been explained by Sabin (100) as due to "constitutional barriers" These are postulated to be anatomical or physiological barriers which develop with advancing age and which prevent certain viruses from progressing either to or within the CNS (barriers at the myoneural junction, at the blood-brain barrier, etc.) The precise nature or role of such constitutional barriers, as well as the significance of nutritional or hormonal factors, remains a matter of speculation

Dissemination of Virus in the Host Organism

The pathogenesis of the viral encephalitides can be separated into two phases: a) an extraneural, systemic infection, b) the encephalitic phase. During the former, varying amounts of virus are demonstrable in the blood stream and there is good experimental evidence to suggest that the arthropod-borne encephalitis viruses multiply in several extraneural organs and tissues. In mice inoculated peripherally with 10–100 lethal doses of Russian spring-summer encephalitis virus, large amounts can be recovered

from the peripheral blood for nine days even though none may be demonstrable in the brain until the fifth day after inoculation (Schlesinger, un-

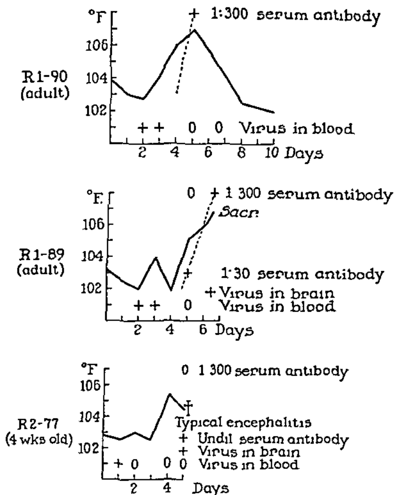


Fig. 3 Effects of Subcutaneous Inoculation of Western Equine Encephalomyelitis Virus in Adult and Young Rabbits (See Text)
(From Schlesinger et al , J Am Med. Assn , 119: 618–620, 1942)

published experiments) This suggests very brisk extraneural multiplication which apparently overwhelms any incipient immune response, and this in turn may explain why adult mice are so highly susceptible to infection with Russian virus by peripheral routes. In infection with the other

viruses, viremia is usually much more transient, and the amount of virus in extraneural organs remains relatively low. As to the mechanism by which virus penetrates from blood into the CNS, several possibilities have been discussed Among the alternatives are: a) progression along peripheral neural pathways, b) spread through perineural lymphatics, c) active growth of the virus through capillary walls, d) migration from capillaries through intercellular spaces, e) deposition of the virus from the blood stream on the nasal mucosa with subsequent invasion by way of the olfactory nerves. Some of these alternatives have been discussed recently by Hurst (59). It may be safe to assume that none of them operates to the exclusion of the others.

HUMAN INFECTION AND IMMUNITY; LABORATORY INFECTIONS

In man, infection with the arthropod-borne encephalitis viruses appears to follow the patterns just described for experimental infection. The systemic and encephalitic phases of infection are clearly separated, and there is ample evidence that the variables discussed play an equally important role However, *the occurrence of laboratory infections with these viruses* in man under circumstances which make arthropod-transmission extremely unlikely leaves little doubt that alternative modes of infection exist. (Eastern E E.—Olitsky and Morgan (89); Western E.E —Fothergill et al. (34), Helwig (52), Gold and Hampil (39), Venezuelan E.E.—Casals et al. (15), Lennette and Koprowski (71), Koprowski and Cox (68); RSSE—Silber and Soloviev (119); Louping ill—Rivers and Schwentker (99), St. Louis—von Magnus (75).) In most cases, precise information as to time or nature of exposure is not available In the cases of Western equine encephalitis described by Helwig and by Gold and Hampil, onset of symptoms followed a known laboratory accident by 14 and 5 days respectively. In the latter, virus was splattered into the patient's eye which may account for the very short incubation period. Helwig's patient was splattered with virus from head to foot, the exact means of entry being unknown. In most cases it is likely that infection occurred by inhalation of finely dispersed infectious material

Thus, while it is certain that some mechanism other than arthropod-transmission *can* bring about encephalitis in man, the evidence in favor of the latter as the common natural mode is overwhelming.

Treatment of Arthropod-Borne Encephalitis

There is no specific therapy for the arthropod-borne encephalitides. Administration of specific hyperimmune serum has been recommended (145) but experimental findings of others (90) make its efficacy in human infection extremely questionable Large amounts used in two cases of laboratory

infection with Western equine virus (39, 52) did not appear to be adequate. Russian workers have reported on the use of hyperimmune serum in Far Eastern encephalitis, with variable and equivocal results (119). Other reliable reports on serotherapy in human cases are not available.

Only supporting and symptomatic measures can be recommended These vary depending on the severity of the case.

NOURISHMENT. Adequate intake of fluids must be insured, if necessary by intravenous drip (1,000 to 3,000 ml of 5 per cent glucose in saline and perhaps 500 ml of 5 per cent amino acids per day).

SEDATION. In excited or delirious or convulsive patients, barbituric acid derivatives, paraldehyde or chloral hydrate should be used Opiates should be avoided

OXYGEN has been described by Wyatt (144) as a most valuable therapeutic adjunct since many of the severest clinical signs appear to be associated with anoxia

The therapeutic efficacy of repeated spinal taps, often mentioned as a measure to relieve headache, is to be doubted Chemotherapeutic agents and antibiotics are without effect in the treatment of encephalitis, but in severe cases their use may be recommended as an aid against complicating infections

Otherwise, the treatment has to be limited to good nursing care, including maintenance of adequate urinary and fecal elimination. Exercises, physiotherapy, and psychiatric help may be required in convalescence

Control Measures

GENERAL MEASURES

Isolation of Cases

Infection by contact or through fomites appears to be so rare that isolation of the patient may be considered a luxury

Vector Control

In endemic areas, all buildings should be provided with adequate screens The success of general eradication measures depends on the particular locale involved, and the advisability of instituting a large-scale program depends on the magnitude of the public health hazard To quote Reeves (97): "A primary consideration in this program will be economic feasi- [illegible]

such a program is in effect, it may be modified to include special [illegible] on vector species . A basic part of any well organized vector control program is modification of the environment to render it unsuitable for the

vector". If a situation arises which makes such measures desirable, competent advice must be sought from entomologists and public health officials.

SPECIFIC IMMUNIZATION

Vaccines have been prepared for all types of arthropod-borne encephalitis. They consist of virus-infected tissue extracts, usually mouse brain or chick embryo, which have been rendered non-infectious by treatment with formaldehyde, ultraviolet light or some other agent. All of these vaccines have been found to be highly effective in protecting experimental animals against infection. Furthermore, in the case of North American equine encephalitides, there is convincing evidence for the efficacy of vaccines in the reduction of infection rates among horses (82). In man, *only* two types of vaccine have been used on a large scale. Smorodintsev (132) reports on favorable results with formalin-inactivated mouse brain vaccine for Russian spring-summer encephalitis. The Japanese encephalitis mouse brain vaccine prepared by Sabin and co-workers (107), or its modification prepared from infected chick embryos (137), have been inoculated into several hundred thousands of people in the Far East (38, *101*, 108, *136*). Very few, if any, instances of severe allergic or neurological side reactions were seen even on repeated vaccination. The question as to the prophylactic efficacy of vaccination cannot as yet be answered satisfactorily. Some cases have occurred in Americans who had either a full or an incomplete course of immunization (101, 136, 144) Wyatt's data, in particular, suggest the possibility that the death rate may be lowered by adequate dosage of vaccine. However, the data thus far available cannot be evaluated because the published reports have not related the case incidence to the total non-vaccinated or vaccinated population Nor is it certain that all the lots of Japanese encephalitis vaccine were fully potent when used in the field

The occurrence of a few cases in immunized people should not discourage further use of vaccines in circumstances which warrant it Vaccination of human beings should be limited to laboratory workers and to people living in or moving into areas of known high epidemic prevalence

Specific Diagnostic Methods—Their Use and Interpretation

The diagnosis of the seasonal, arthropod-borne encephalitides has to be made by laboratory tests These consist of a) virus isolation, b) serological tests The technical aspects of these procedures have been reviewed in a number of recent papers (17, 43).

VIRUS ISOLATION

Indications

Theoretically, a laboratory equipped for diagnostic virus studies can, if provided with the proper clinical and pathological specimens, confirm or

exclude an infection with most of the viruses now known or thought to be directly encephalitogenic. In practice, however, because of the prohibitive number of agents, it is almost impossible to include all of them in routine tests. This applies especially to attempts to isolate virus from patients. Such an undertaking is always a time-consuming research project. Success depends upon the selection of test materials at the proper time and upon their proper treatment. The laboratory worker should be familiar with the clinical, pathological, and epidemiological aspects of the case in order to select the most promising approach. He is aware that his knowledge of the characteristics of *known* viruses may be of no service in the identification of a newly isolated variety. Hence, even if standardized procedures for isolation and identification of recognized encephalitogenic viruses fail, his curiosity will lead him to try different laboratory animals or variations in other steps. Because of these ramifications, virus isolation from suspected cases is really too elaborate a procedure to be practiced routinely in sporadic cases. It should be reserved for exceptional situations or for investigation of epidemics

Technique

The "standard" procedure for isolation of an arthropod-borne encephalitis virus is, briefly, the following: a) suitable test material, e g, spinal fluid or brain tissue extract, is inoculated into laboratory animals, preferably infant or weanling mice and usually by the intracerebral route, b) if, after several days, the test animals show signs of illness, they are sacrificed, and their brain is transferred to other animals of the same species to initiate serial passages, c) presence of bacteria is excluded, d) standardized immune sera are used in attempts to identify the newly recovered agent by comparison with stock laboratory strains of known viruses; e) serological tests are done with the new strain and the serum of the donor patient in order to establish that the virus really came from him and was not "picked up" in the course of laboratory manipulations.

Further aids in the classification of newly isolated strains of virus are: f) their physical and chemical characteristics (such as filterability, size, and sensitivity to physical and chemical agents); g) their experimental host range.

Characteristics of the Viruses

The size of all known arthropod-borne encephalitis viruses is in the small and intermediate range on the scale of recognized viruses, i e, 15 to 50 mμ. Thus, they pass through the common bacteria-retaining filters and remain, in significant concentration, in the supernatant after centrifugation for 1 hour at 18,000 rpm. They are not affected by chemotherapeutic agents or

antibiotics, but are inactivated by ether, oxidizing agents and formaldehyde, and by exposure to x-rays or ultraviolet light. Each virus has its own optimal pH range for maximum activity, although this may depend partly on the type of medium in which each virus is being tested.

In properly buffered media, all these viruses retain their activity for months or years at freezing temperatures or in the lyophilized state. 50 per cent buffered glycerol will also protect their activity for years at 4°C.

Experimental Host Range

The summer encephalitis viruses are pathogenic for a large variety of experimental animals other than the mouse. Chick embryos, rabbits, guinea pigs, hamsters, cotton rats, and monkeys are the most commonly used species. Characteristic differences exist, not only in host range of newly isolated strains, but also in regard to a) adaptability to a variety of hosts on continued passage, b) infectivity by different routes of inoculation, c) infectivity for members of the same species of varying ages, d) the type and the pathogenesis of the disease induced in experimental animals.

These differences, important though they may be to the laboratory worker, are relevant as an illustration of the many steps involved in the characterization of a new strain of virus If, regardless of the complexities of virus isolation procedures, it is decided to go ahead with such attempts, success depends on the selection of proper specimens and on their reaching the laboratory in suitable condition

Specimens Suitable for Isolation of Viruses

The most promising source of virus is *CNS tissue* obtained at autopsy of rapidly fatal cases If death is delayed until more than five to ten days after onset, demonstrable virus is apt to have disappeared

Spinal Fluid yields virus in rare instances of arthropod-borne encephalitis, and then only if it is obtained very early in the acute phase. In this respect, these maladies differ from lymphocytic choriomeningitis in which virus persists in the spinal fluid for long periods

Blood or Serum The isolation of virus from blood or serum has been reported on rare occasions (SLE—Blattner and Heys (7), WEE—Howitt (55); Jap. E—Kobayashi (66); RSSE—Silber and Soloviev (119). This again is a mark of distinction from lymphocytic choriomeningitis in which the virus remains in peripheral blood during most of the febrile stage.

Oral or Naso-pharyngeal Washings have yielded virus in laboratory infections with Venezuelan equine encephalitis virus (15, 68, 71), perhaps because the mode of infection probably was not the bite of infected mosquitoes. There is nothing to suggest that secretions of the upper respiratory or alimentary tract, or urine, or feces are promising test materials in naturally occurring cases

Storage and Shipment of Specimens to Be Used for Virus Isolation

CNS Tissue Necropsy material should be obtained under aseptic precautions as soon after death as possible, and before embalming. Blocks of about 1 ccm. each should be taken from various areas of the cortex, including hippocampus, from the midbrain, thalamus, pons, medulla, cerebellum, and various levels of the spinal cord. *They must not be fixed.* If the laboratory is in the immediate vicinity, the specimens should be taken there in a suitable, sterile container and without delay. If the material has to be in transit for more than a few minutes, it must be kept cold. It may be frozen in a dry ice box If this is not possible, it should be placed in a thermos jug with chopped ice, care being taken that no water gets into the vessel containing the tissue. The choice of suitable containers for the specimen should be dictated by concern for the safety of laboratory workers, since serious laboratory infections with the majority of the arthropod-borne viruses have been reported. Leakage on the outside of the containers must be avoided. Most ordinary glass will crack in an atmosphere of dry ice or when it is thawed. Therefore plastic tubes with screw caps ("Lusteroid") are most convenient for frozen samples The next best thing is a large, wide-mouthed, *thick-walled glass bottle tightly closed with a rubber stopper which should* be secured with adhesive tape If prolonged storage is unavoidable, or dry ice or mechanical freezing equipment not available, the method of choice is refrigeration of the tissue in autoclaved 50 per cent glycerol in buffered saline.

Unless serum has been obtained from the patient shortly before death, blood should be collected from the heart at autopsy. It is not unusual to find antibody in the serum at a stage of the illness at which virus is present in the CNS. The demonstration of antibody in the serum may be valuable corroboration of the verdict that the virus was actually recovered from the CNS and was not a contaminant.

Spinal Fluid to be used for virus studies had best be preserved frozen in a sealed glass ampoule

Blood intended for virus isolation should be collected in a dry sterile syringe either without anticoagulant or with heparin Citrate and oxalate are toxic when inoculated intracerebrally into experimental animals and should not be used as anticoagulants Plasma or serum should be separated from the cells as soon as possible and refrigerated or preferably frozen in sealed glass ampoules Red cells or clots may be sent to the laboratory in the frozen state There is no reason to believe that the resulting hemolysis would damage virus adsorbed on the cells However, separation of serum prior to freezing is essential, because hemolysis would ruin it for confirmatory serological tests.

IMMUNOLOGICAL PROCEDURES

Methods and Techniques

Regardless of whether or not virus isolation is resorted to in diagnosis, identification of the etiological agent depends on immunological tests. The following methods are available: a) active immunity tests, b) passive immunity tests, c) neutralization tests, d) complement-fixation tests, and e) hemagglutination-inhibition tests, for certain viruses only. Other serological methods, such as precipitin, agglutinin and flocculation reactions, have not been developed for the encephalitic viruses.

ACTIVE IMMUNITY TESTS. Graded amounts of a newly isolated virus are inoculated into groups of experimental animals previously actively immunized against a number of known reference strains, or the latter strains are used to challenge animals immunized with the unknown agent Resistance to challenge suggests a relationship between the immunizing and the challenge strain. On the other hand, absence of resistance does not necessarily rule out a relationship because under certain experimental conditions two immunologically identical strains of the same virus may be far apart in their ability to reveal a given degree of immunity. Therefore, active immunity tests often are not as sensitive as other methods If positive, they may be more specific than neutralization tests.

PASSIVE IMMUNITY TESTS. The relationship between the unknown and known strains is established by challenging animals previously inoculated with hyperimmune serum, usually obtained by immunizing rabbits. This technique has the disadvantage of being relatively insensitive, large doses of potent serum being required to obtain significant degrees of resistance On the other hand, certain non-specific factors present in serum which may give apparent cross reactions in neutralization tests may be eliminated by giving immune serum and challenge virus separately and by different routes of inoculation.

NEUTRALIZATION TEST. This technique is used for the identification of newly isolated viruses as well as for the serological diagnosis of cases from which virus has not been isolated, and for epidemiological survey studies Depending on the virus used and on the sensitivity required, the test can be carried out in two ways: either undiluted serum (immune) is mixed with graded amounts of virus, or a standard amount of virus (10 to 100 minimal lethal doses) is mixed with varying dilutions of test serum After incubation at 37°C for two hours, each mixture is inoculated into suitable test animals. Depending on the technique used, the results can be expressed either in terms of the number of lethal doses of virus neutralized ("neutralization index") or of the highest serum dilution inactivating a standard

dose of virus ("neutralizing titer"). There are certain advantages to either technique which are fully discussed in technical reviews (17, 43, 86). Another important variable is the choice of test animal and of the route of inoculation. All arthropod-borne encephalitis viruses are highly pathogenic for adult mice, causing a fatal disease after intracerebral inoculation. For most neutralization tests, this method is the simplest and is sufficiently sensitive. There are, however, exceptions Mixtures of Russian spring-summer encephalitis virus and potent immune sera may remain infectious when inoculated intracerebrally and yet prove inert when injected intraperitoneally in mice of the same breed and age (16). With other neurotropic viruses which are not highly invasive by the intraperitoneal route in adult mice, one may have to substitute 7 to 14-day-old mice in order to increase the sensitivity of neutralization tests This variation may reveal significant amounts of antibody even in sera in which none is demonstrable by the intracerebral route (based on findings by Olitsky and Harford (88)).

This difference in sensitivity between intracerebral and intraperitoneal neutralization tests is of interest not only because of its implications in regard to diagnostic accuracy but also as a clue to the pathogenesis of experimental infection with these and other neurotropic viruses It is likely that under the conditions applying in most neutralization tests, the virus-antibody mixtures contain an excess of free antibody as well as some incompletely neutralized virus particles If such a mixture is placed within immediate reach of selectively susceptible cells, i e, within the CNS, a single infectious unit can presumably initiate infection. If the same mixture is inoculated into a more remote site, free antibody is available to check the wide dissemination of virus particles, and the chances of un-neutralized particles gaining a foothold in the CNS are considerably reduced. Regardless of the mechanism, there have been several instances in which the choice of the most sensitive technique available had a decisive influence on the outcome of the diagnostic tests.

COMPLEMENT-FIXATION TEST. The principle of this method does not differ from complement-fixation reactions with other viral or non-viral systems. *The antigens are specific and are derived from tissues infected* with each particular virus *Methods for their preparation and detailed* descriptions of the technique of the test will again be found in several pertinent papers (14, 17, 43). *This test, to a greater extent* than the other immunological and serological methods, reveals certain real or apparent cross relationships among some of the arthropod-borne encephalitis viruses and between members of this group and of other virus families (13, 72, 104, 125). Despite this overlapping, it is the most useful diagnostic tool and a highly sensitive one Many of the arthropod-borne encephalitis and other virus antigens are now commercially available, placing this technique

at the disposal of any diagnostic laboratory equipped to carry out Wassermann tests.

HEMAGGLUTINATION INHIBITION. Of the arthropod-borne encephalitis viruses, only three—Japanese, Russian spring-summer, and St. Louis encephalitis viruses—are capable of agglutinating *in vitro* erythrocytes of certain mammalian and avian species (106). Of the suspected agents, West Nile virus and the viruses of the Mengo encephalitis group have similar properties Specific inhibition of hemagglutination by homologous immune sera has been demonstrated. The technique is still impractical and too limited for routine use in diagnostic laboratories.

Selection and Treatment of Specimens for Serological Diagnostic Tests

The production of antibody begins early in the acute stage of the illness. Antibody may persist for many years after recovery or after a silent infection, and therefore the unequivocal diagnosis of a given case depends, not on the demonstration of its presence, but on the demonstration that it is newly formed or increases in the course of the illness under investigation. It is mandatory that at least two samples of serum be submitted to the diagnostic laboratory, one collected as soon after onset as is practicable, and the second one to two weeks later. These samples must be tested simultaneously. Therefore, a definitive diagnosis cannot be made until the disease is well along on its course. At least 10 ml of blood are collected under aseptic conditions with a dry syringe The blood is allowed to clot, the serum is separated and sent to the laboratory with as little delay as possible Freezing of the serum is very desirable. If facilities for freezing are not available, the serum must be kept under constant refrigeration *Whole blood must not be frozen* since the resulting hemolysis renders the specimen unusable for complement-fixation tests No preservatives, anti-bacterial agents, or anticoagulants should be added to the blood or serum. The serum should not be heat-inactivated before reaching the laboratory The laboratory should insist that each specimen be accompanied by a resumé of the patient's record, with special emphasis on a) date of onset and pertinent anamnestic data, b) character of the illness, c) clinical laboratory findings, d) clinical diagnoses Specimens submitted without these data or with a request "for virus study" without any specification should be rejected Single specimens should be accepted and tested only under special circumstances and at the discretion of the laboratory.

ILLUSTRATION OF THE PRACTICAL APPLICATION OF "STANDARD" DIAGNOSTIC PROCEDURES

The following example is cited to illustrate the application and interpretation of the laboratory methods just outlined:

During the summer of 1946, laboratory studies were carried out on a small outbreak of Japanese encephalitis among American soldiers stationed in an isolated camp in Southern Korea. There were three cases of encephalitis in a total camp population of 1500 (111). The first patient suffered a rapidly progressing, fatal illness. Onset of prodromal symptoms was on August 21, onset of definite encephalitic signs on August 24. He died during the night of the 26th. Necropsy was performed in Korea on the 27th, and amply refrigerated fragments of CNS tissue reached the Tokyo laboratory in the morning of August 28, along with a sample of serum obtained twelve hours before death. Table 9 illustrates the outcome of three serial passages in mice initiated with CNS material from this case.

TABLE 9

Isolation of "Korea" Virus from Brain and Cord of Fatal Case 1 and Passage in Mice

PASSAGE	INOCULUM	RESULT OF MOUSE INOCULATION*
1	Human cerebral cortex, cerebellum, medulla and spinal cord, 30 per cent suspension in physiological salt solution, 0 02 ml intracerebrally and 0 2 ml intra-abdominally into 8 to 10 day old mice	5, 5, 5, 5, 5, 6, 6, 6, 6
2	10 per cent mouse-brain suspension, 0 03 ml intracerebrally into old mice	4, 4, 4, 4, 4, 5
3	20 per cent mouse-brain suspension; 0 03 ml intracerebrally into old mice	4, 4, 4, 4, 4, 4

* Numbers in this column refer to days after inoculation when each mouse first exhibited signs of involvement of the central nervous system

From Sabin, Schlesinger, Ginder and Matumoto Am J Hyg , 46: 356, 1947

Brains of mice showing encephalitic signs on the fifth day after inoculation of the original material proved to be free of bacteria They were used for preparation of antigen for a complement-fixation test with standard immune sera. Comparison of this antigen with a stock strain of Japanese encephalitis and control antigens proved the identity of the new strain with the former (table 10).

Thus, the nature of the virus presumably isolated from the patient was established on the fifth day after material had been received at the laboratory. Confirmation of the identification consisted of a) neutralization of the newly isolated strain by a standard Japanese encephalitis immune rabbit serum (table 11), b) demonstration of neutralizing antibody in the serum of the donor patient taken twelve hours before death It neutralized not only the newly isolated virus but also a standard strain of Japanese encephalitis virus (table 12) This test also illustrates the marked difference in sensitivity between the intracerebral and the intraperitoneal method—a

technical point of the greatest practical significance in cases of this sort, especially if the result of the intracerebral test is entirely negative or

TABLE 10

*Identification of the "Korea" Virus as Japanese B Virus by Complement-Fixation Test**

Known Immune Serum		Complement Fixation with Indicated Antigens				
Type	Original dilution†	Korea	Jap B	SLE	WEE	Saline
Japanese B (Jap B)	4	4‡	4			0
	8	4	4			
	16	4	4			
	32	4	4			
	64	3	4			
	128	2	4			
	256	1	3			
	512	1	1			
St Louis (SLE)	4	0	0	4		0
	8	0	0	4		
	16	0	0	3		
	32	0	0	2		
	64	0		1		
	128	0		±		
	256	0		0		
	512	0		0		
Western equine (WEE)	2	0	0	0	4	0
	4	0	0	0	4	
	8	0			4	
	16	0			4	
	32				4	
	64				3	
	128				±	
	256				0	

* 2.5 exact units of complement were used in this test as determined by simultaneous titration of the complement in the presence of each of the antigens used

† Dilution expressed as 4 = 1 in 4, as 8 = 1 in 8, etc

‡ The numerals indicate the extent of fixation, 4 representing complete fixation and 0, complete hemolysis, or no fixation The titer of a serum is the original dilution added to the mixture which gives 2 plus (approximately 50%) or better fixation

From Sabin, Schlesinger, Ginder and Matumoto Am J Hyg, 46: 356, 1947

equivocal The serum of Case 1 failed to fix complement in the presence of Japanese encephalitis antigen

Two additional cases of severe but non-fatal encephalitis had their onset

within six days of the first one. Diagnosis in these two cases was based on the results of the complement-fixation tests shown in table 13. Sera from both patients were obtained on the fourth day after onset. At that time, Case 2 had already demonstrable antibody reacting specifically with Japanese encephalitis antigen. Three days later, the titer was four-fold higher, enough to consider concurrent infection with this virus as proved beyond reasonable doubt. Had the seventh day serum been the earliest one in this case, no increase could have been shown and the evidence would have been less convincing. The fourth day serum of Case 3, on the other hand, was negative, while later samples reacted to high titer, again indicating antibody production resulting from concurrent infection.

TABLE 11

Identification of "Korea" Virus as Japanese B Virus by Intracerebral Neutralization Test in Mice

SERUM	10^{-2}	10^{-3}	10^{-4}	10^{-5}	10^{-6}	10^{-7}	10^{-8}	LD_{50} (LOG)	NEUTRAL. INDEX*
Normal rabbit control	4/4†	4/4	4/4	4/4	4/4	3/4	3/4	8 2+?	—
Japanese B immune rabbit	4/4	3/3	1/4	0/4	0/4	0/4	—	3.7	32,000+?

* Neutralization index titer of virus in normal control serum divided by the titer of virus in test serum.

† Numerator: number of mice succumbed, denominator: number of mice inoculated.

From Sabin, Schlesinger, Ginder and Matumoto. Am. J. Hyg , *46: 356, 1947.*

SCOPE AND LIMITATIONS OF SPECIFIC DIAGNOSTIC TESTS

Evaluation of Positive or Negative Findings in the Study of Epidemics and in Endemic Areas

The Korean episode has been presented in some detail because it exemplifies several points of general validity for all known forms of arthropod-borne encephalitis:

(1) The identification of a case by means of virus isolation and serological tests can be accomplished in a few days.

(2) Even under the most favorable conditions, the specific laboratory diagnosis comes too late to affect the outcome of the case

(3) The recovery of virus from a fatal case presents no difficulties if it is there. However, *failure to recover virus has no diagnostic significance*, especially if death occurred relatively late There are on record several cases of human as well as of experimental infection in which the specific etiology could be confirmed serologically (*i.e*, *by an increase in antibody*) even though the CNS was free of virus at the time of death.

TABLE 12

Neutralizing Antibodies in Serum of Fatal Case 1 of Japanese B Encephalitis 5 Days After Appearance of First Systemic Symptom

Virus Used	Type of Test	Serum	Dilution of Virus in Mixtures 10^{-2}	10^{-3}	10^{-4}	10^{-5}	10^{-6}	10^{-7}	10^{-8}	10^{-9}	LD_{50} (Reciprocal of Log of Dilution)	Neutralization Index
Japanese B Nakayama strain	Intracerebral* (4-week-old mice) 0.03 ml	Control†	—	—	—	5/5	5/5	5/5	0/5	—	7.5	—
		Case 1	5/5	5/5	5/5	0/5	0/5	0/5	—	—	4.5	1,000
	Intraperitoneal (2-week-old mice) 0.1 ml	Control†	—	—	5/5	5/5	5/5	5/5	3/4	1/4	8.5	—
		Case 1	0/4	0/4	0/4	0/4	0/5	0/4	—	—	1.5–?	10,000,000+?
"Korea" strain from case 1	Intracerebral	Control†	—	—	—	—	5/5	3/3	5/5	3/5	9.2+?	—
		Case 1	5/5	5/5	5/5	1/5	1/5	0/4	—	—	4.7	32,000+?

* The intraperitoneal and intracerebral tests were carried out simultaneously, the same serum-virus mixtures being used.

† Normal rabbit.

From Sabin, Schlesinger, Ginder, and Matumoto Am. J. Hyg. **46**, 356, 1947.

(4) Presence of specific neutralizing or complement-fixing antibody is acceptable as evidence that the patient has been, *at one time or another*, exposed to and infected with the corresponding virus or one closely related to it, but it does not necessarily signify recent or concurrent infection

(5) *Identification of the current or recent illness depends on the demonstration of a rise* in antibody titer in the course of the illness.

(6) *Demonstrability of neutralizing antibody in significant amounts may* depend, especially in early sera, on the use of methods more sensitive than the standard intracerebral neutralization test.

(7) Neutralizing antibody usually persists for many years, apparently without the necessity of repeated infection Complement-fixing antibody,

TABLE 13

Specific Serologic Diagnosis of Nonfatal Cases by Complement-Fixation Test

CASE	DAYS AFTER ONSET WHEN SERUM OBTAINED	ANTIGEN ADDED TO INDICATED DILUTION OF SERUM (1)										
		Japanese encephalitis						SLE		WEE		Saline
		2	4	8	16	32	64	2	4	2	4	2
2	4	3	2	1	+			0	0	0	0	0
	7	4	4	4	2			0	0	0	0	0
	15	4	4	4	3	1	0	0	0	0	0	0
3	4	0	0	0	0			0	0	0	0	0
	13	4	4	4	3			0	0	0	0	0
	20	4	4	4	3			0	0	0	0	0

Table abbreviated from Sabin, Schlesinger, Ginder and Matumoto Am J Hyg, 46: 356, 1947.

on the other hand, may decline after a few weeks or months, hence *its* presence is more indicative of *recent* infection. This is of the greatest practical importance, especially when it comes to the identification of cases in endemic areas where the majority of the population may have acquired neutralizing antibody largely as a result of wholesale nonapparent and perhaps repeated infection Here, it is the complement-fixation rather than the neutralization test which may reveal changes in the antibody pattern In the event that such changes cannot be shown, the suspected virus, while not ruled out conclusively, cannot be incriminated as the one responsible for the outbreak under investigation Since the arthropod-borne encephalitides are so highly endemic, it is precisely this sort of situation which presents the most frequent challenge to the efficacy and usefulness of serological methods.

This last point is illustrated by a report on an alleged outbreak of Jap-

anese encephalitis in Korea in the summer of 1949 involving 5,548 native cases with 2,429 deaths (58). The incrimination of Japanese encephalitis virus was based on a single confirmed instance of isolation from human CNS tissue. Six additional isolations, reportedly successful in the Laboratories of Seoul National University, could not be repeated by the authors using the same autopsy materials (instances of "false" isolations of Japanese encephalitis virus, due to laboratory contamination, have been reported by Sabin (105)). In addition, the following serological evidence was presented: a) encephalitis patients: all of 12 single convalescent serum specimens contained neutralizing antibody; only four of 17 tested had complement-fixing antibody. b) "Normal" Koreans: 17 of 19 tested had neutralizing antibody of high titer, and one of 20 gave a positive complement-fixation test. This is not a significant difference between the two groups. While one might assume that both groups had acquired their antibodies recently, the low incidence of complement-fixing antibody notwithstanding, available evidence does not support this assumption. In 1946, three years before the 1949 epidemic, samples of the same "normal" population, i.e. people living in and around Seoul, and of the same age group, had been tested by Deuel et al. (22) for serological evidence of nonapparent infection with Japanese encephalitis virus. At that time 80 per cent of the sera tested contained neutralizing antibody. Thus, while the virus undoubtedly was causing some cases in Korea in 1949, the evidence is against rather than for the conclusion that the majority or all of the more than 5,000 cases were part of a single epidemic due to this agent. Questioning this conclusion is more than raising a merely hypothetical issue. The unequivocal demonstration that a virus can raise such havoc in a population thoroughly immune to it through natural exposure would obviously be of the most far-reaching significance. Among other things, it would present the most damaging argument against the usefulness of specific prophylactic vaccination. Moreover, the definitive identification of so large a number of cases on such tenuous grounds would forestall any effort to search for other etiological factors. It would add thousands of case records to the already confused picture of the Japanese and other forms of arthropod-borne encephalitis. Finally, it would have profound effects on statistics and measures relating to public health.

Reluctance to accept the evidence presented in this instance as adequate is borne out by Sabin's investigation of 38 cases of suspected Japanese encephalitis among Americans on Okinawa in 1945 (101) to which reference has already been made. This series is particularly instructive because it dealt with a previously non-exposed group suddenly brought into an endemic focus and into immediate proximity to an outbreak among the natives. In this environment it was only natural that all patients

acute neurological signs were considered victims of the same illness. This seemed justified by the clinical similarity between the twelve in whom the diagnosis could be confirmed serologically and some of the 26 on whom serological tests were negative, even on specimens collected relatively late in convalescence. Some of the latter suffered from mild illnesses of the "aseptic meningitis" variety, and only one of them had clinically typical encephalitis. Poliomyelitis could not be ruled out for some of the cases. Sabin writes: "It is especially noteworthy that . . . it was not possible to prove infection by the Japanese B encephalitis virus in patients who exhibited only nuchal rigidity, transitory reflex changes and pleocytosis as the only manifestation indicative of involvement of the nervous system—the syndrome ordinarily described as aseptic or lymphocytic meningitis, and, when it is seen during the course of a poliomyelitis epidemic, as nonparalytic poliomyelitis. . . *There was no evidence that during this outbreak* (on Okinawa) it represented a mild form of encephalitis due to Japanese B virus. However, in patients who exhibited this syndrome plus a certain amount of drowsiness, lethargy, and mental confusion, positive serological evidence . . . was obtained". This latter conclusion was later confirmed by the observations of others in 1947–1949 (136)

The experience of those who have carried out extensive diagnostic studies in areas in which other arthropod-borne encephalitides are endemic has been similar. Hammon and Reeves (49) published the following figures for the incidence of serologically confirmed St. Louis and Western equine encephalitis among 44 patients believed to have had neurotropic virus infections in Kern County, California, in 1944.

St Louis	3
Western equine encephalitis	2
Paralytic poliomyelitis	4
Encephalitis of unknown etiology or non-paralytic poliomyelitis	35

It may be argued that infection with the seasonal encephalitis virus may take place without resulting in detectable antibody formation, but this has never been shown unequivocally One should not hesitate to accept serological evidence as the most sensitive and, indeed, as the only reliable diagnostic tool short of virus isolation

Obviously, it is impossible to do laboratory tests on every case in a large epidemic Valid conclusions can be reached on the basis of representative samples, provided the number is significant and the types of specimens permit conclusive tests If these conditions are fulfilled, and the results still do not prove decisive, it is better to leave the etiology or the homogeneity of the epidemic in the realm of uncertainty Serological methods, if used and interpreted correctly, have their chief usefulness in the investigation of focal outbreaks and in epidemiological surveys

Efficacy and Usefulness of Specific Diagnostic Tests in Non-endemic Regions and in Identification of Sporadic Cases

The previous section served to show the difficulty encountered in trying to separate a specific type of encephalitis from unrelated diseases even within a localized outbreak or a known endemic focus. When it comes to an evaluation of the role played by the summer encephalitis and the other readily identifiable encephalitogenic viruses in the causation of sporadic cases, the result is highly disappointing The vast majority of cases

TABLE 14

Summary of Positive Findings in Complement-Fixation Tests on Cases of Suspected Virus Infections of the CNS During First 30 Months of Operation, Virus Diagnostic Laboratory of the Department of Health, City of New York

CLINICAL DIAGNOSIS OR TEST REQUESTED	NUMBER OF CASES	NUMBER CONFIRMED AS			NUMBER OF POSITIVE TESTS
		Mumps	Lymphocyt choriomen	Lympho-gran venereum	
"Virus encephalitis or meningitis"	111	8	3	1*	12
"Mumps encephalitis"	13	9			9
"Virus studies"	21	6			6
"Lymphocytic choriomeningitis"	15	1	1		2
"Lymphocytic choriomeningitis or mumps"	2	2			2
Total	162	26†	4	1	31
Per cent	100	16	2 5	0 6	19.1

* This patient had inguinal lymphadenopathy but a negative Frei test when submitted for diagnosis

† It is probable that about 50 per cent of these patients had concurrent parotitis.

which, on clinical grounds, justify a diagnosis of "probable viral infection of the CNS" other than paralytic poliomyelitis are not due to one of these agents This conclusion is based on the findings of several virus diagnostic laboratories in different sections of the United States In table 14, a summary is given of the experience of the virus diagnostic laboratory of the Department of Health of the City of New York during the first 30 months of operation from 1948 to 1950 * Most of the specimens were submitted from major teaching hospitals in the metropolitan area, and the tentative diagnoses and requested tests were based in most cases on careful clinical and laboratory studies Many of the specimens, especially those which

* These data were collected and made available to the author by Miss Olga Simonovic of the Bureau of Laboratories, Department of Health, City of New York Her assistance is gratefully acknowledged.

came with requests for "encephalitis tests" or unspecified "virus studies", were tested for rises in complement-fixing antibody against all or most of the following viruses: Eastern and Western equine, St. Louis, Japanese, lymphocytic choriomeningitis, mumps, lymphogranuloma inguinale, and rabies Of the 162 cases, only 31 (19 per cent) could be diagnosed by specific serological tests. Of these, 26 (84 per cent) were due to mumps virus. Since in this group probably about one-half of the patients had concurrent parotitis, the percentage of diagnosed cases should be corrected to consider only those with purely neurological manifestations, and this would leave about 11 per cent of the total The only other viruses which could be incriminated were those of lymphocytic choriomeningitis (2 5 per cent) and lymphogranuloma inguinale (1 case). Not a single case of arthropod-borne encephalitis could be identified in this random group. Even if one assumes—and this is a legitimate assumption—that a fair number of these cases might have had non-paralytic forms of poliomyelitis or of Coxsackie virus infections, there still remains a sizable number whose cause is beyond our present routine diagnostic skill.

That this New York experience is not isolated, is illustrated by data from the Department of Virus and Rickettsial Diseases, Army Medical Department Research and Graduate School, which were compiled by Dr. Ross Gauld and kindly made available by him and Dr J. E Smadel They represent the results of diagnostic tests done in that laboratory during the periods of July 1946 to June 1947 and January to December 1948 The material differs from that of the New York laboratory in that it consists of specimens from members of the Armed Forces in various parts of the world, probably including some of the areas in which one or another of the seasonal encephalitides is endemic Of a total of 401 cases of "neurotropic virus disease" registered during the two periods, 60 (15 per cent) yielded positive tests, as follows·

"Viral encephalitis"	
Japanese	3
Western equine	2
St Louis	1
Venezuelan equine	1 (laboratory infection)
Russian spring summer	1 (laboratory infection)
Rabies	1
	9 (2 2%)
"Aseptic meningitis"	
Lymphocytic choriomeningitis	9 (2 2%)
Mumps	42 (10 5%)
	51 (12 7%)

It is striking that these results, obtained from so widely separate sources, should be so similar to those obtained in New York Information from other diagnostic laboratories is entirely in line with these data.

It is clear, then, that the arthropod-borne encephalitis viruses play a very minor role in non-epidemic meningo-encephalitis of suspected viral etiology. Despite the tremendous strides made during the past two decades in the classification and characterization of virus infections of the CNS, many forms—like von Economo's encephalitis—still belong in the category "etiology unknown". This point must be stressed, because it is important for clinicians and laboratory workers to be constantly mindful of the present limitations of our knowledge as well as of its rapidly increasing scope.

BIBLIOGRAPHY

1 ADAMSON, J D AND DUBO, S . Clinical findings in encephalitis (Western equine) Canad J Pub Health, 33: 288-300, 1942

2 AFZELIUS-ALM, L Aseptic (nonbacterial) encephalomeningitides in Gothenborg 1932-1950 Acta Med Scand , Suppl 263, 96 pp , 1951.

3 ARMSTRONG, M P , WILSON, F H , MCLEAN, W J AND SILVERTHORNE, N Studies on poliomyelitis in Ontario II Isolation of the Coxsackie virus in association with poliomyelitis virus A preliminary report Canad J Pub Health, 41: 51-59, 1950

4 AYRES, J C AND FEEMSTER, R F The sequelae of Eastern equine encephalomyelitis New Eng J Med , 240: 960-962, 1949

5 BAKER, A B AND NORAN, H H Western variety of equine encephalitis in man A clinicopathologic study Arch Neurol & Psychiat , 47: 565-587, 1942

6 BAWELL, M B , DEUEL, R E , JR , MATUMOTO, M AND SABIN, A B Status and significance of inapparent infection with virus of Japanese B encephalitis in Japan in 1946 Am J Hyg , 51: 1-12, 1950

7 BLATTNER, R J AND HEYS, F M. Isolation of St. Louis encephalitis from the peripheral blood of a human subject J Ped , 28: 401-405, 1946

8 BLATTNER, R J AND HEYS, F M Blood-sucking vectors of encephalitis experimental transmission of St Louis encephalitis (Hubbard strain) to white Swiss mice by American dog tick Dermacentor variabilis Say J Exp Med , 79, 439-454, 1944

9 BODIAN, D Poliomyelitis Neuropathologic observations in relation to motor symptoms J A M A , 134. 1148-1154, 1947

10 BURNS, K F , TIGERTT, W D AND MATUMOTO, M Japanese equine encephalomyelitis 1947 epizootic II Serological and etiological studies Am J Hyg , 50. 27-45, 1949

11 BURNS, K F Congenital Japanese encephalitis infection of swine Proc Soc Exp Biol & Med , 75. 621-625, 1950

12 BUSS, W C AND HOWITT, B F Human equine encephalomyelitis in Kern County, California, 1938, 1939 and 1940 Am J Pub Health, 31: 935-944, 1941

13 CASALS, J Immunological relationships among central nervous system viruses J Exp Med , 79. 341-359, 1944

14 CASALS, J Complement-fixation test for diagnosis of human viral encephalitides J Immunol , 56. 337-341, 1947

15. Casals, J., Curnen, E. C and Thomas, L.: Venzuelan equine encephalomyelitis in man J. Exp Med., 77: 521–530, 1943
16 Casals, J. and Olitsky, P K.: Enduring immunity following vaccination of mice with formalin-inactivated virus of Russian spring-summer (Far Eastern, tick-borne) encephalitis. J Exp. Med , 82: 431–443, 1945.
17 Casals, J and Olitsky, P. K. The diagnosis of neurotropic virus infections, including the viral encephalitides, lymphocytic choriomeningitis, and poliomyelitis In "Diagnosis of viral and rickettsial infections", F. L Horsfall, Jr , Editor. New York, Columbia University Press, 1949.
18 Casals, J. and Webster, L T : Close relation between Russian spring-summer encephalitis and louping-ill viruses Science, 97: 246–248, 1943
19 Cleland, J B , Campbell, A W and Bradley, B.: The Australian epidemics of an acute polioencephalomyelitis (X-disease). Annual Rep , Microbiol Lab of the Dept of Publ. Health, Sydney, N.S.W., 1917.
20 Dalldorf, G and Sickles, G M : An unidentified filtrable agent isolated from the feces of children with paralysis Science, 108: 61–62, 1948
21 Davison, G , Neubauer, C. and Hurst, E W Meningo encephalitis in man due to the louping-ill virus Lancet, 255: 453–457, 1948.
22 Deuel, R E , Jr , Bawell, M B , Matumoto, M. and Sabin, A B : Status and significance of inapparent infection with virus of Japanese B encephalitis in Korea and Okinawa in 1946 Am J Hyg , 51: 13–20, 1950
23 Dick, G W A The relationship of Mengo encephalomyelitis, encephalomyocarditis, Columbia-SK and M M. viruses J Immunol , 62: 375–386, 1949
24 Dick, G W A , Best, A M , Haddow, A J and Smithburn, K C Mengo encephalomyelitis A hitherto unknown virus affecting man Lancet, 255: 286–289, 1948
25 Dick, G W A and Haddow, A J · Uganda S virus In press
26 Donovan, C R and Bowman, M Some epidemiological features of poliomyelitis and encephalitis, Manitoba, 1941 Canad J Pub Health, 33: 246–257, 1942
27 Duffy, C E Interference between St Louis encephalitis virus and equine encephalomyelitis virus (Western type) in the chick embryo Science, 99: 517–518, 1944
28 Durand, P Virus filtrant pathogène pour l'homme et les animaux de laboratoire, et à affinités méningée et pulmonaire Arch Inst Pasteur de Tunis, 29: 179–227, 1940
29 Eklund, C M Human encephalitis of Western equine type in Minnesota in 1941, clinical and epidemiological study of serologically positive cases Am J Hyg , 43: 171–193, 1946
30 Enders, J F , Weller, T H and Robbins, J F Cultivation of Lansing strain of poliomyelitis virus in cultures of various human embryonic tissues Science, 109: 85–87, 1949
31 Farber, S , Hill, A , Connerly, M L and Dingle, J H Encephalitis in infants and children caused by the virus of the Eastern variety of equine encephalitis J. A M A , 114: 1725–1731, 1940
32 Feemster, R F Outbreak of encephalitis in man due to the Eastern virus of equine encephalomyelitis Am J Pub Health, 28: 1403–1410, 1938
33 Findlay, G. M Durand's Disease A virus infection transmissible to animals and man Trans Roy Soc Trop Med & Hyg , 35: 303–318, 1942
34 Fothergill, L D , Holden, M and Wyckoff, R W G. Western equine encephalomyelitis in a laboratory worker J A M A , 113: 206–207, 1939

35 FREUND, J · Accumulation of antibodies in the central nervous system. J. Exp Med, 51: 889–902, 1930.
36. GARD, S. AND HELLER, L. Hemagglutination by Col-MM-virus. Proc. Soc Exp. Biol & Med, 76: 68–73, 1951.
37 GETTING, V A: Equine encephalomyelitis in Massachusetts An analysis of the 1938 outbreak, a follow-up of cases and a report of a mosquito survey New Eng J. Med, 224: 999–1006, 1941
38 GINDER, D. R, MATUMOTO, M, SCHLESINGER, R. W. AND SABIN, A B.: Neutralizing and complement-fixing antibodies for Japanese B encephalitis virus in vaccinated U. S. personnel in Japan Proc. Soc. Exp. Biol. & Med, 65: 130–135, 1947.
39 GOLD, H. AND HAMPIL, B Equine encephalomyelitis in a laboratory technician with recovery Ann. Int. Med, 16: 556–569, 1942
40 HAMMON, W. McD.· Encephalitis in the Yakima Valley. Mixed St. Louis and Western equine types J A. M. A., 117: 161–167, 1941.
41. HAMMON, W. McD.· Encephalitis Eastern and Western equine and St. Louis types as observed in 1941 in Washington, Arizona, New Mexico and Texas J A. M. A, 121: 560–566, 1943
42. HAMMON, W. McD · *Arthropod-borne virus encephalitides (Charles Franklin* Craig Lecture, 1947). Am J. Trop Med, 28: 515–525, 1948
43 HAMMON, W McD.· Encephalitis In "Diagnostic procedures for virus and rickettsial diseases". Am Pub. Health Assn, 1st ed, p 187–217, 1948
44 HAMMON, W McD · Public health problems relating to the viral encephalitides in the Far East and the Pacific Islands Proc 17th Ann Conf, Calif Mosquito Control Assn., p 13–15, 1949.
45 HAMMON, W McD, LUNDY, H W, GRAY, J A, EVANS, F C, BANG, F AND IZUMI, E M. A large-scale serum neutralization survey of certain vertebrates as part of an epidemiological study of encephalitis of the Western equine and St. Louis types J Immunol, 44: 75–86, 1942
46 HAMMON, W. McD. AND REEVES, W C · Western equine encephalomyelitis virus in the blood of experimentally inoculated chickens J Exp Med., 83: 163–173, 1946
47 HAMMON, W McD, REEVES, W C AND IZUMI, E St. Louis encephalitis virus in the blood of experimentally inoculated fowls and mammals J Exp Med, 83: 175–183, 1946
48 HAMMON, W McD, REEVES, W C AND BURROUGHS, R Japanese B encephalitis virus in the blood of experimentally inoculated chickens Proc Soc Exp Biol. & Med, 61· 304–308, 1946
49 HAMMON, W. McD AND REEVES, W C Interepidemic studies on Arthropod-borne virus encephalitides in Kern County, California and Yakima Valley, Washington, 1944 Am J. Hyg, 46: 326–335, 1947
50 HAYASHI, M.· Übertragung des Virus von Encephalitis epidemica auf Affen Proc. Imp Acad Tokyo, 10: 41–44, 1934
51 HAYMAKER, W AND SABIN, A B. Topographic distribution of lesions in central nervous system in Japanese B encephalitis, nature of lesions, case on Okinawa Arch Neurol & Psychiat, 57: 673–692, 1947
52 HELWIG, F C. Western equine encephalomyelitis following accidental inoculation with chick embryo virus J A M A, 115: 291–292, 1940
53 HELWIG, F C AND SCHMIDT, E C H A filter-passing agent producing interstitial myocarditis in anthropoid apes and small animals Science, 102: 31–33, 1945.

54. HOWITT, B. F.: Recovery of the virus of equine encephalomyelitis from the brain of a child. Science, 88: 455–456, 1938
55. HOWITT, B. F.: Recovery of virus (Western type) from human blood serum (equine encephalomyelitis). Science, 89: 541-542, 1939.
56. HOWITT, B F : *Development of neutralizing antibodies to the viruses of equine* encephalomyelitis (Western strain) and St. Louis encephalitis in the blood and cerebrospinal fluid of man and animals, together with recovery of the St Louis virus from the blood of monkeys J Immunol., 42: 117-131, 1941.
57. HUANG, C H . A visible method for titration and neutralization of viruses on the basis of pH changes in tissue cultures Proc Soc. Exp Biol & Med , 54: 158-160, 1943
58. HULLINGHORST, R L , BURNS, K. F , CHOI, Y. T. AND WHATLEY, L R.: Japanese B encephalitis in Korea The epidemic of 1949 J A. M A , 145: 460–466, 1951
59. HURST, E W Some observations on the pathogenesis of Eastern equine encephalomyelitis and louping-ill in young and old animals, with special reference to routes of entry of the viruses into the nervous system J Comp Path & Ther , 60: 237–262, 1950
60 JUNGEBLUT, C W., SANDERS, M AND FEINER, R R.: Studies in rodent poliomyelitis I Further experiments with the murine strain of SK poliomyelitis virus J Exp Med , 76: 611–629, 1942
61. JUNGEBLUT, C W AND DALLDORF, G : Epidemiological and experimental observations on the possible significance of rodents in a suburban epidemic of poliomyelitis Am J Pub Health, 33: 169–172, 1943
62 JUNGEBLUT, C W AND HORVATH, B Inhibition of Columbia SK virus hemagglutination by polio-convalescent human sera Fed Proc , 10: 411, 1951
63 KABAT, E A , MOORE, D H. AND LANDOW, H An electrophoretic study of the protein components in cerebrospinal fluid and their relationship to the serum proteins J Clin Invest , 21: 571–577, 1942
64 KANEKO, R AND AOKI, Y Ueber die Encephalitis epidemica in Japan Erg Inn Med und Kinderheilkunde, 34: 342–356, 1928
65 KASAHARA, S , UEDA, M , OKAMOTO, Y , YOSHIDA, S , HAMANO, R AND YAMADA, R Experimental studies on epidemic encephalitis Transmission test of Japanese encephalitis in 1935 and some characteristics of infectious agents Kitasato Arch Exp Med , 13: 48–65, 1936
66 KOBAYASHI, R Comparison of concentration of virus of Japanese encephalitis in blood, cerebrospinal fluid and brain tissues of encephalitis patients and monkeys and mice infected with the virus Kitasato Arch Exp Med , 17: 45–52, 1940
67 KOPROWSKI, H AND COX, H R Adaptation of Colorado tick fever virus to mouse and developing chick embryos Proc Soc Exp Biol & Med , 62. 320–322, 1946
68 KOPROWSKI, H AND COX, H R Human laboratory infections with Venezuelan equine encephalomyelitis virus Report of four cases New Eng J Med , 236: 647–654, 1947
69. LAEMMERT, H. W., JR AND HUGHES, T P Virus of Ilhéus encephalitis, isolation, serologic specificity and transmission J Immunol , 55: 61–67, 1947
70 LEAKE, J. P : Epidemic of infectious encephalitis Pub Health Rep , 56: 1902–1905, 1941.
71. LENNETTE, E H AND KOPROWSKI, H Human infection with Venezuelan equine encephalomyelitis virus Report on eight cases of infection acquired in the laboratory. J A M A , 123: 1088–1095, 1943

72 LENNETTE, E. H. AND KOPROWSKI, H · Antigenic relationships of West Nile, Japanese B encephalitis and St Louis encephalitis viruses J Immunol, 52: 235-246, 1946
73. LEWIS, L, TAYLOR, H. G, SOREM, M B, NORCROSS, J. W. AND KINDSVATTER, V. H.. Japanese B encephalitis. Clinical observations in an outbreak on Okinawa Shima Arch Neurol. & Psychiat, 57: 430-463, 1947.
74. MACKENZIE, R. D AND FINDLAY, G. M · The production of a neurotropic strain of Rift Valley Fever virus Lancet, 230: 140-141, 1936
75 VON MAGNUS, H.· Laboratory infection with St. Louis encephalitis virus Acta pathol. Scand, 27: 276-282, 1950
76 Matheson Commission Epidemic encephalitis, New York, Columbia Univ. Press, 1929.
77. Matheson Commission· Epidemic encephalitis Third report. New York, Columbia Univ Press, 1939
78 McCORDOCK, H. A, COLLIER, W. AND GRAY, S H.· The pathologic changes of the St Louis type of acute encephalitis J A. M A, 103: 822-825, 1934
79 MEDOVY, H . Western equine encephalomyelitis in infants J Ped, 22: 308-318, 1943
80 MELNICK, J L · Epidemic of poliomyelitis characterized by dual infections with Coxsackie and poliomyelitis viruses Fed Proc, 10: 415-416, 1951.
81 MEYER, K F., HARING, C M AND HOWITT, B. F.· Newer knowledge of neurotropic virus infections of horse J. Am. Vet M. A, 79: 376-389, 1931.
82. MILLER, A. W Report on infectious equine encephalomyelitis in United States in 1944 J. Am Vet M A, 107: 10-13, 1945.
83 MORGAN, I M Influence of age on susceptibility and on immune response of mice to Eastern equine encephalomyelitis virus J Exp Med, 74: 115-132, 1941.
84 MORGAN, I M, SCHLESINGER, R W AND OLITSKY, P. K · Induced resistance of the central nervous system to experimental infection with equine encephalomyelitis virus I Neutralizing antibody in the central nervous system in relation to cerebral resistance J Exp Med., 76: 357-369, 1942
85 MUCKENFUSS, R S, ARMSTRONG, C AND McCORDOCK, H A : Encephalitis, studies on experimental transmission Pub Health Rep, 48: 1341-1343, 1933
86 OLITSKY, P K AND CASALS, J Neutralization tests for diagnosis of human virus encephalitides J A. M A, 134: 1224-1228, 1947
87 OLITSKY, P K AND CASALS, J. Viral encephalitides In "Viral and Rickettsial Infections of Man" Th M Rivers, ed J B Lippincott Co, Publishers, pp 163-212, 1948
88 OLITSKY, P. K AND HARFORD, C. G Intraperitoneal and intracerebral routes in serum protection tests with the virus of equine encephalomyelitis. I. A comparison of two routes in protection tests J Exp Med, 68· 173-189, 1938
89 OLITSKY, P. K. AND MORGAN, I M Protective antibodies against Eastern equine encephalomyelitis virus in the serum of laboratory workers Proc. Soc Exp Biol & Med, 41: 212-215, 1939
90 OLITSKY, P K, SCHLESINGER, R W AND MORGAN, I M Induced resistance of the central nervous system to experimental infection with equine encephalomyelitis virus II Serotherapy in Western virus infection. J Exp Med, 77: 359-374, 1943
91 PERDRAU, J R The Australian epidemic of encephalomyelitis (X-disease) J Path & Bact, 42: 59-65, 1936
92. POND, W L, WARREN, J. AND RUSS, S B A serological study of strains of

Russian spring-summer encephalitis and louping-ill viruses. Bact Proc, p 83–84, 1951.

93 Pool, W A, Brownlee, A. and Wilson, R D: The etiology of "louping ill" *J Comp. Path & Therap*, *43: 253–290, 1930.*

94 Public Health Bulletin No. 214 · Report on the St Louis outbreak of encephalitis U. S. Government Printing Office, Washington, 1935.

95. Quong, T. L: Pathology of Western equine encephalomyelitis, 18 human cases, Manitoba epidemic, 1941, Canad. J Pub Health, **33:** 300–306, 1942

96 Randall, R and Mills, J W.: Fatal encephalitis in man due to the Venzuelan virus of equine encephalomyelitis in Trinidad Science, **99:** 225–226, 1944

97 Reeves, W. C. The encephalitis problem in United States Am J Pub Health, **41:** 678–686, 1951

98 Reeves, W C and Hammon, W. McD. Feeding habits of the proven and possible mosquito vectors of Western equine and St Louis encephalitis in the Yakima Valley, Washington Am J Trop Med, **24:** 131–134, 1944

99 Rivers, T M and Schwentker, F. F.. Louping-ill in man J Exp Med, **59:** 669–685, 1934

100 Sabin, A B Constitutional barriers to involvement of the nervous system by certain viruses, with special reference to the role of nutrition. J. Ped, **19:** 596–607, 1941

101 Sabin, A B Epidemic encephalitis in military personnel, isolation of Japanese B virus on Okinawa in 1945, serologic diagnosis, clinical manifestations, epidemiologic aspects and use of mouse brain vaccine J. A M. A, **133:** 281–293, 1947

102 Sabin, A B "Dengue" in Viral and Rickettsial Infections of Man, edited by T. M Rivers J B Lippincott Company, pp 445–453, 1948

103 Sabin, A B Viral infections of the human nervous system Classification and general considerations Proc 4th Intern Neurol Congress, Paris, pp 85–94, 1949

104. Sabin, A B Antigenic relationship of dengue and yellow fever viruses with those of West-Nile and Japanese B encephalitis Fed Proc, **8:** 410, 1949.

105 Sabin, A. B Search for virus of Japanese B encephalitis in various arthropods *collected in Japan in 1946–1947 Am J Hyg*, *51: 36–62, 1950*

106 Sabin, A B and Buescher, E L. Unique physico-chemical properties of Japanese B encephalitis virus hemagglutinin Proc Soc Exp Biol & Med, **74:** 222–230, 1950

107 Sabin, A B, Duffy, C E, Warren, J, Ward, R, Peck, J L and Ruchman, I · The St Louis and Japanese B types of epidemic encephalitis Development of noninfective vaccines report of basic data J A M A, **122:** 477–486, 1943

108 Sabin, A B, Ginder, D R, Matumoto, M and Schlesinger, R W Serological response of Japanese children and old people to Japanese B encephalitis mouse brain vaccine Proc Soc Exp Biol & Med, **65:** 135–140, 1947

109. Sabin, A B, *Ginder*, D R *and Matumoto*, M *Difference in dissemination* of the virus of Japanese B encephalitis among domestic animals and human [illegible]–355, 1947

[illegible] Production of immunity to dengue with [illegible]ice Science, **101:** 640–642, 1945.

[illegible] Japanese [illegible] –375, 1947

[illegible] Okayama

in relation to the epidemiology of Japanese B encephalitis Am. J. Hyg., 51: 21-35, 1950.
113 SCHLESINGER, R W.: The mechanism of active cerebral immunity to equine encephalomyelitis virus I Influence of the rate of viral multiplication. J. Exp. Med, 89: 491-505, 1949
114 SCHLESINGER, R. W.. The mechanism of active cerebral immunity to equine encephalomyelitis virus. II The local antigenic booster effect of the challenge inoculum J Exp. Med, 89: 507-527, 1949
115 SCHLESINGER, R. W: Propagation in chick embryos of dengue virus Hawaiian strain II Findings in infected eggs Proc. Soc. Exp Biol & Med, 76: 817-823, 1951
116 SCHLESINGER, R. W, MORGAN, I. M. AND OLITSKY, P. K.: Significance of neutralizing antibody in experimental equine encephalomyelitis A consideration of its relation to the disease in man: preliminary report. J A. M. A., 119: 618-620, 1942
117. SCHLESINGER, R. W., OLITSKY, P K. AND MORGAN, I M.: Observations on acquired cellular resistance to equine encephalomyelitis virus Proc Soc Exp Biol & Med, 54: 272-273, 1943.
118 SCHLESINGER, R W., OLITSKY, P. K. AND MORGAN, I M.: Induced resistance of the central nervous system to experimental infection with equine encephalomyelitis virus III. Abortive infection with Western virus and subsequent interference with the action of heterologous viruses J. Exp Med, 80: 197-211, 1943
118A. SHAW, E W., MELNICK, J L AND CURNEN, E. C: Infection of laboratory workers with Coxsackie viruses Ann. Int. Med, 33: 32-40, 1950
119 SILBER, L A AND SOLOVIEV, V D Far Eastern tick-borne spring-summer (spring) encephalitis. Am. Rev Soviet. Med, Special Suppl, 1946
120 SIMPSON, T W AND MEIKLEJOHN, G: Sequelae of Japanese B encephalitis Am J Trop Med, 27: 727-731, 1947.
121 SMADEL, J E: Research in virus diseases Bull U. S Army Med Dept, 7: 795-808, 1946
122. SMITH, M, BLATTNER, R J. AND HEYS, F M St Louis encephalitis infection of chicken mites, *Dermanyssus gallinae*, by feeding on chickens with viremia, transovarian passage of virus into the second generation J Exp Med, 84: 1-6, 1946
123 SMITH, M G, BLATTNER, R J AND HEYS, F. M St Louis encephalitis Transmission of virus to chickens by infected mites *Dermanyssus gallinae* and resulting viremia as source of virus for infection of mites J. Exp Med, 86: 229-237, 1947.
124 SMITH, M G, BLATTNER, R J, HEYS, F M AND MILLER, A: Experiments on the role of the chicken mite, *Dermanyssus gallinae*, and the mosquito in the epidemiology of St Louis encephalitis J Exp Med, 87: 119-138, 1948.
125 SMITHBURN, K C.: Differentiation of West-Nile virus from viruses of St Louis and Japanese B encephalitis J Immunol, 44: 25-31, 1942
126 SMITHBURN, K C AND HADDOW, A J Semliki forest virus I. Isolation and pathogenic properties. J Immunol, 49: 141-157, 1944
127 SMITHBURN, K C AND HADDOW, A J.. Ntaya virus A hitherto unknown agent isolated from mosquitoes collected in Uganda Proc Soc Exp. Biol & Med., 77: 130-133, 1951.
128 SMITHBURN, K C, HADDOW, A J AND MAHAFFY, A. F: A neurotropic virus

isolated from *Aedes* mosquitoes caught in the Semliki forest. Am J Trop Med., 26: 189–208, 1946

129. SMITHBURN, K. C, HUGHES, T. P., BURKE, A. W AND PAUL, J H. A neurotropic virus isolated from the blood of a native of Uganda Am. J. Trop. Med, 20: 471–492, 1940.

130 SMITHBURN, K. C., MAHAFFY, A. F AND PAUL, J H: Bwamba fever and its causative virus Am. J. Trop Med, 21: 75–90, 1941.

131 SMORODINTSEV, A. A.: The spring-summer tick-borne encephalitis Arch f d ges Virusforschg, 1: 468–480, 1940

132 SMORODINTSEV, A. A. Tick-borne encephalitis. Am Rev Sov Med., 1: 400–408, 1944.

133 SYVERTON, J T. AND BERRY, G. P.: Hereditary transmission of the Western type of equine encephalomyelitis virus in the wood tick, *Dermacentor Andersoni Stiles* J. Exp Med, 73: 507–529, 1941

134 TENBROECK, C. AND MERRILL, M H Serological difference between Eastern and Western equine encephalomyelitis virus. Proc Soc Exp Biol. & Med, 31: 217–220, 1933

135. THEILER, M. Studies on the action of yellow fever virus in mice. Ann Trop Med & Parasit, 24: 249–272, 1930

136 TIGERTT, W D AND HAMMON, W McD, with coworkers Japanese B encephalitis: A complete review of experience on Okinawa 1945-1949. Am J Trop Med, 30: 689–722, 1950

137 WARREN, J AND HOUGH, R G. A vaccine against Japanese B encephalitis prepared from infected chick embryo Proc Soc Exp Biol & Med, 61: 109–113, 1946

138 WARREN, J, RUSS, S B AND JEFFRIES, H Neutralizing antibody against viruses of encephalomyocarditis (EMC) group in sera of wild rats Proc Soc Exp Biol & Med, 71: 376–378, 1949

139 WARREN, J, SMADEL, J E AND RUSS, S. B The family relationship of encephalomyocarditis, Columbia-SK, M M and Mengo encephalomyelitis viruses J Immunol, 62: 387–398, 1949

140 WEBSTER, L T Japanese B encephalitis virus Its differentiation from the St Louis encephalitis and relationship to louping-ill virus J Exp Med, 67: 609–618, 1938

141 WEBSTER, L T AND CLOW, A D Experimental encephalitis (St Louis type) in mice with high inborn resistance A chronic subclinical infection J Exp Med, 63: 827–845, 1936

142 WEBSTER, L T AND FITE, G L A virus encountered in the study of encephalitis in the St Louis and Kansas City epidemics of 1933 Science, 78: 463–465, 1933

143 WEBSTER, L T AND FITE, G L St Louis encephalitis, serological relation to Japanese encephalitis and experimental studies on immunity Science, 79: 254–255, 1934

144 WYATT, N F · Japanese B encephalitis Report of five cases J Lab & Clin Med, 34: 1656–1670, 1949

145 ZICHIS, J AND SHAUGHNESSY, H J Successful treatment of experimental Western equine encephalomyelitis with hyperimmune rabbit serum Am J Pub Health, 35: 815–823, 1945

146 ZIMMERMAN, H M The pathology of Japanese B encephalitis Am J Path, 22: 965–991, 1946

Sickle Cell Anemia

BYRD S LEAVELL, M.D AND WILLIAM A. MacILWAINE, M.D.

INTRODUCTION

When the preparation of this article was begun, there was no recent comprehensive review or complete bibliography on sickle cell anemia despite the marked increase in interest and the significant advances in knowledge of many aspects of the disease that have occurred in recent years. When our manuscript was in its final phase of revision, the comprehensive review by Margolies (275) appeared Although the concept of the two articles was evidently similar and much of the same literature was covered by both, the treatment of the material and the points of emphasis in the discussions are different

We have made an effort to include in the bibliography all articles published between 1910 and the end of 1951, but undoubtedly unintentional omissions have occurred.

NOMENCLATURE

"Sickle cell anemia" (278) is the name used most often for the disease characterized by a varied clinical picture, a hemolytic type of anemia, and erythrocytes that assume a bizarre "sickle-shape" under conditions of low oxygen tension "Sickle cell trait" (171), "Sicklemia" (99), "latent sickling" (418), and "sacklemia" (99) are terms that have been suggested to indicate those individuals who are free from the clinical manifestations of sickle cell anemia although their erythrocytes have the sickling characteristic. The term "sickle cell disease" (30) was prompted by the concept that the sickling trait and sickle cell anemia are extremes of the same fundamental disorder which might produce symptoms even in the absence of anemia Other terms that have been proposed are "meniscocytosis" (158, 159), "drepanocyte", "drepanocytemia", and "drepanocytic anemia" (172) The expression "latent sickle cell anemia" has been used to indicate sickle cell trait and also mild degrees of sickle cell anemia The Committee for Clarification of the Nomenclature of Cells and Diseases of the Blood and Blood Forming Organs has recommended the terms "sicklemia without anemia" (trait) and "sicklemia with normocytic anemia" (90).

HISTORICAL

To Dr. James B Herrick belongs the credit for reporting the first case of sickle cell anemia in 1910 In an article entitled "Peculiar Elongated and

Sickle-Shaped Red Blood Corpuscles in a Case of Severe Anemia" (195), he reported the case of a 20-year-old Negro male student from Grenada, West Indies. This patient consulted him because of a respiratory infection and fever, but also had what is now recognized as the classical picture of sickle cell anemia. Herrick noted "we were at a loss to account for this peculiar complexus of symptoms, a condition evidently chronic as revealed by the history of the past three years, with yaws and suppurative otitis as predecessors, yet with acute exacerbations, a condition not clearly explained on the basis of an organic lesion in any organ, yet showing cardiac enlargement, albuminuria and cylinduria, general adenopathy, icterus, with a secondary anemia not remarkable for the great reduction of red corpuscles or hemoglobin, but strikingly atypical in the number of nucleated red corpuscles of the normoblastic type and in the tendency of the erythrocytes to assume a slender sickle-like state. The leukocytosis with a rather high eosinophile count is also to be noted." In this noteworthy paper, Herrick gave a clear description of the clinical manifestations of the disease, noted its chronic nature, and reported the essential and characteristic abnormality of the blood He also stated, "Whether the blood picture represents merely a freakish poikilocytosis or is dependent on some peculiar physical or chemical condition of the blood, or is characteristic of some particular disease, I cannot at present answer." It is interesting that at the present time it appears that all the possibilities which Herrick mentioned have been established The blood picture does present a "freakish poikilocytosis" which is thought to be due to a "peculiar" chemical condition of the hemoglobin, and it is considered to be characteristic of a "particular disease"

Prior to Herrick's report, others had seen sickled erythrocytes and sickle cell anemia but failed to appreciate the significance. In 1911, Washburn (449) reported the second case of sickle cell anemia. This patient had been under observation for several years before Herrick's report, and in 1908, Dr. John Staige Davis, late Professor of Medicine at the University of Virginia, made the following note on her chart, "Many normoblasts and most peculiar poikilocytes, being scythe-shaped" (258) It is possible that others had observed these forms even earlier In 1889, Hayem (187) described "semilunar" erythrocytes which he interpreted as artifacts In 1904, Dresbach (130) reported "elliptical" shaped erythrocytes, usually considered as distinct from sickle forms, in a mulatto student who suffered from rheumatic symptoms and died a year later in cardiac failure, but no association was made between the abnormal erythrocytes and the clinical findings "Demilune" erythrocytes were noted in the blood of an Algerian with malaria by Sergent and Sergent in 1905 (373).

DISTRIBUTION

Geography

Sickle cell disease probably occurs wherever members or descendants of the Negro race are found. In the continental United States, where the majority of the case reports have originated, the disorder appears to be more frequent in the Southern states where the Negro population is greater, but it is limited to no section Outside of the United States the largest numbers of case reports have come from locations where the dark races predominate. Africa (140, 459, 18, 10, 473, 34, 33, 315), Central America (427, 74, 295, 4, 335, 479), Cuba (9, 84, 473, 178), and South America (369, 370, 371, 269) have been represented by a number of case reports.

Race

In 1922, Mason first noted that sickle cell anemia appeared to be a dis-

and Houseal concluded that the disorder occurred only in Negroes (370). In 1927, it was stated "the presence or absence of sicklemic trait, because of its high incidence in the Negro race, may be of some service in certain medico-legal procedures; for example, the determination of paternity as a supplement to the use of present knowledge in the realm of hemagglutinins" (37). In 1943, Odgen reported two cases of Nicaraguan origin and made the following statement. "I assert, without hesitation, that the presence of the sickling trait in a white person is definite proof of admixture of Negro blood in the immediate ancestry" (309).

The first instance of sickle cell anemia in a white individual was reported by Castana in 1925 (75). A review of the literature has revealed reports of 36 additional cases These are listed in table 1. The "nationalities" represented by white persons with sickle cell anemia include English, Scotch, Irish, Jewish, Spanish, Cuban, Greek, Italian, Native Sicilian, Russian-Jewish, German, Indian and "American".

At least 12 of the 36 case reports listed in table 1 seem to be instances of definite sickle cell anemia in an individual who would be accepted as white by ordinary criteria (100, 96, 129, 163, 273, 290, 351). Most striking of these have been two white boys of Native Greek parents (100) and a 9-year-old boy of Italian extraction (351) The family tree of this latter individual was traced back five generations without the discovery of any evidence which would suggest any admixture of Negro blood. Three genera-

TABLE 1

Summary of Reported Cases of Sickle Cell Anemia in White Individuals

CASE	REFERENCE	DATE	SEX	AGE	COLOR	BIRTH	FATHER	MOTHER
1	(75)	1925	M		W*	Italy		
2	(18)	1925	M		B*		Arabian	Arabian
3	(410)	1927	F	6	O*	U S	English-Jewish	Cuban
4	(243)	1927	F	32	W	U S.	Scotch-Irish	Spanish
5	(100)	1929	M	4	W	U S.	Native Greek	Native Greek
6	(100)	1929	M	14	W	U. S	Native Greek	Native Greek
7	(244)	1931	M	64	W	†	†	†
8	(381)	1931	M	48	W	U. S.	Scotch-Irish	Scotch-Irish
9	(351)	1932	M	9	W	U S.	Native Italian	Native Italian
10	(86)	1933	M	3	W	U S.	Native Sicilian	Native Sicilian
11	(86)	1933	M	11	W	U. S.	Native Sicilian	Native Sicilian
12	(327)	1934	F	30	W	U S	Native Russian Jewish	Native Russian Jewish
13	(96)	1934	M	10 mo	W	U S.	Native "American"	Native "American"
14	(96)	1934	F	3½	W	U S.	Native "American"	Native "American"
15	(81)	1937	M	10	†	†	†	†
16	(215)	1937	M	5	W	†	Greek	†
17	(455)	1937	M	8	W	U S	Sicilian	Sicilian
18	(455)	1937	M	—	W	U S	Sicilian	Sicilian
19	(170)	1937	F	8	W	Sicily	Sicilian	Sicilian
20	(170)	1937	F	15	W	Sicily	Sicilian	Sicilian
21	(331)	1939	F	13	W	Sicily	Native Sicilian	Native Sicilian
22	(117)	1940	F	22	W	U S	Native Italian	Native Italian
23	(117)	1940	F	9	W	U S	Native Italian	Native Italian
24	(273)	1941	F	20	W	†	†	†
25	(440)	1941	M	6	W	U S	Greek	Greek
26	(446)	1941	F	49	W	†	Native Greek	Native Greek
27	(290)	1942	F	4	W	†	Native Italian	Native Italian
28	(290)	1942	F	1½	W	†	Native Italian	Native Italian
29	(309)	1943	F	9	W	U S	Native Nicaraguan (Spanish)	Native Nicaraguan (Spanish)
30	(309)	1943	F	8	W	U S	German	? Indian-Scotch
31	(163)	1943	F	54	W	Italy	Unknown	Unknown
32	(475)	1945	M	20	W	†	†	†
33	(475)	1945	F	14	W	†	†	†
34	(129)	1948	M	3	W	U S	Native Italian	Native Italian
35	(332)	1950	M	38	W	U S	Sicilian American	Native Sicilian
36	(156)	1951	M	26	W	Pres U S	†	†
37	(162)	1951	M	22	W	Greece	Greek	Greek

* W, white, B, brown, O, octoroon

† Not mentioned.

tions of this child's family exhibited the sickling trait. The majority of the white patients that have been reported thus far are of Mediterranean origin Because of the proximity of these countries to Africa, the intermingling of the black and white races in the past might explain the occurrence of sickle cell anemia in some Italians, Sicilians, and Greeks of the present day. Sicily was once a province of Carthage when there was free social intermingling of Italians, Greeks and Africans on that island (86) Another possible explanation of the increased frequency of sickle cell anemia in white individuals with Mediterranean ancestry is given in the interesting report of Powell, Rodarte and Neel (412), who reported sickle cell anemia in a male whose father had the sickling trait while the mother had thalassemia minor.

Several studies have reported that the incidence of sicklemia is higher in individuals who are mainly Negro, but who also have Caucasian or American Indian ancestry, than it is in full-blooded Negroes (122, 199). In the latter report on 60 persons with sicklemia, both the physical characteristics and genealogic history were studied. It was considered that a significantly lower incidence occurred in the pure-blooded Negro and "7/8 Negro" as compared to those with 5/8 and 9/8 fractions. Further study is necessary before this question can be answered

In summary both sickle cell trait and sickle cell anemia appear to be primarily disorders of the Negro race which may occur in a lower incidence in other races. The preponderance of Mediterranean ancestry in the white cases reported and the apparent relationship of the sickling trait to the genes strongly suggest that admixture of Negro blood occurred at some time in the past, perhaps remote, in families of persons with sicklemia. A similar opinion has been expressed in an editorial review on the subject (135)

Incidence

In 1924, Sydenstricker (415) first estimated that 5 4 per cent of all Negroes had sickle cell anemia In addition, he divided the disorder into active and latent phases, and noted that "the latent phase is much more common of the two, the ratio being nine to one in my series " Other authors have reported studies on the incidence in the American Negro (31, 32, 50, 81, 101, 117, 122, 127, 159, 181, 199, 215, 217, 228, 244, 250, 274, 287, 301, 309, 398, 413, 414, 427, 451, 474), in the white population in the United States (47, 66, 81, 122, 146, 228, 287, 309, 383, 418), in the Africa Negro (11, 35, 138, 139, 146, 247a, 337, 358, 420), and in the Negro in Central and South America (47, 66, 84, 263, 283, 284, 383, 427). In these reports the incidence of sickle cell trait in American Negroes has varied from 3.4 per cent (366) to 14 per cent (414) In addition to the locality, other factors

may be responsible for the marked differences in the results. The age of the group may influence the results, since the incidence appears to be higher in younger individuals than in those over the age of 30 years (201). Furthermore, the method of determining sickling also influences the figures, since some of the new methods are more sensitive than the original coverslip method used in earlier surveys. The results of the reported surveys for sickle cell trait and sickle cell anemia in Negroes and whites in the United States and in other parts of the world are treated in considerable detail in the review by Margolies (275). *The incidence of sickling in the 22,170* Negroes tested in the United States was 9 per cent. It appears that the incidence of sickle cell anemia is about one-fortieth that of the sickle cell trait, about 2.25 per 1,000 Negroes.

Age

Sickle cell anemia is for the most part a disease of young persons. Although it has been observed that the active disease is rarely compatible with life beyond the age of 30 (201), there have been frequent instances of the disorder in individuals past this age In one series of 19 patients, seven were over 30 years of age (143) Individuals who lived 70 (201), 62 and 46 (143), 72 (417), and 78 (415) years have been reported. Probably the youngest case of sickle cell anemia observed thus far is that which occurred in a 19-day-old infant who survived a hemolytic crisis at that age (153) Several other authors have noted the disease in infants during the first year of life (474, 49, 121, 357, 19, 43, 366, 87). Sickle cells have been noted in the blood of babies at birth (415). In a survey of the incidence of sicklemia in newborn infants, Scott, Crawford and Jenkins (366) studied several sicklemic infants in the first five days of life but did not observe true sickle cell anemia in a newly born They found an incidence of 3 4 per cent in newly borns but an incidence of 7.6 per cent in older children and suggested that there is a lower incidence of expression of the sickle cell trait in early life than in later life Watson, Stahman and Billello (451) have suggested that the "relative suppression of sickling phenomenon in the blood of young infants" may be attributed to "the presence of fetal hemoglobin which does not disappear until the age of about 4½ months"

Sex

The reports of the frequency of sicklemia in the two sexes are not in agreement. In the literature prior to 1932, 63 per cent of the reported cases were males (15, 16) In another survey (81) only 30.8 per cent of 120 individuals with sicklemia were males In a survey in Panama, an incidence of 8.6 per cent was found in 335 females compared to 4 1 per cent in 293 males (427). These reported differences in the sexes appear to be due to the

small size of the groups that were studied. Margolies (275) analyzed all of the reported survey studies and found an incidence of 10.98 in 4,909 females and 10 91 in 6,241 males.

MECHANISM OF DISEASE

Heredity

The hereditary nature of the sickling characteristic and sickle cell anemia was first commented upon in 1922 by Mason (278). The theory of a dominant gene was advanced by Huck in 1923 (201) and was accepted for many years. Huck made careful studies, but was handicapped by the use of the older methods of testing for the sickling property which probably gave some false negative reactions. Neel (300, 301) has made an extensive study of the heredity factor in 61 families and concluded that the sickling characteristic is a gene-transmitted disorder, heterozygous in persons with the trait and homozygous in those with anemia. This theory requires that an abnormal sickling gene be present in both parents of patients with sickle cell anemia. Another possible mechanism for the development of sickle cell anemia is given in the report by Powell, Rodarte and Neel (412) of sickle cell anemia in a male whose father had the sickling trait while the mother had thalassemia minor This may afford another explanation of the high incidence of Mediterranean ancestry in the reported instances of sickle cell anemia in Caucasians. Still another genetic background for sickle cell anemia or a disease very similar to it has been reported (224a) Kaplan, Zuelzer and Neel reported a disease that was clinically indistinguishable from sickle cell anemia, in which one parent had the sickle cell trait and the other an abnormal hemoglobin that differed electrophoretically from both normal and sickle cell anemia hemoglobin

Neel's theory, that the sickling gene is heterozygous in individuals with the sickle cell trait and homozygous in those with sickle cell anemia, is in accord with the results obtained in the Negro families we have studied. An example is given in figure 1 Both parents had the sickling trait while the various offspring had normal erythrocytes, or the sickling trait, or sickle cell anemia.

Sickling Property

Although Herrick (195) first described the sickle cell, it remained for Huck (201) to demonstrate that the sickling characteristic resided in the erythrocytes and not in the plasma Hahn and Gillespie (172) showed that the form of the erythrocyte was reversible and depended upon the degree of oxygenation of the hemoglobin, since an atmosphere of oxygen produced the discoid form and the sickle cell shape appeared when the hemoglobin

was reduced. The importance of the hemoglobin in the sickling process was also indicated by Ponder's observation that cells from which the hemoglobin had been removed, so-called "ghost cells", could not be made to sickle (328). Similar evidence was provided by the interesting report of Harris (184a) who demonstrated that stroma-free solutions of deoxygenated hemoglobin prepared from the blood of patients with sickle cell anemia contained tactoids *with shapes similar to sickle cells* when observed with the polarizing microscope.

Even more definitive work on the nature of the hemoglobin defect has

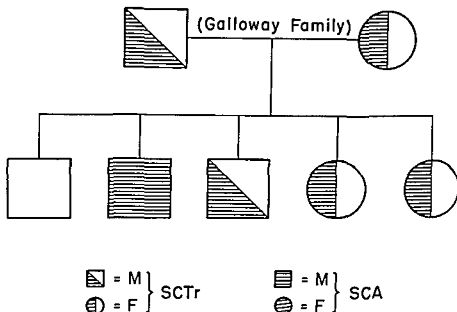


FIG 1 HEREDITY IN SICKLE CELL DISEASE

been reported by Pauling, Itano, Singer, and Wells (317, 318). In some noteworthy experiments these workers demonstrated that hemoglobin obtained from erythrocytes of patients with sickle cell anemia differed electrophoretically from that present in normal cells in that hemoglobin from the two sources acted as differently charged ions under certain conditions They also reported that the hemoglobin of patients with sickle cell trait appeared to be a mixture of these two hemoglobins and observed that in 25 individuals with the sickle cell trait whom they examined, the sickle cell anemia hemoglobin varied from 25 per cent to 44 per cent In both of these reports they indicated their reasons for believing that the abnormality lay in the protein portion of the hemoglobin molecule and might be in the nature of a different physical arrangement of the polypetide chains Wells and Itano (456) demonstrated small quantities of hemoglobin that behaved as

normal hemoglobin electrophoretically in some patients with mild forms of sickle cell anemia. Neel, Wells and Itano (301a) found the abnormal hemoglobin to vary from 22.3 per cent to 45 2 per cent in individuals with sickle cell trait. They concluded that this difference is genetic in origin, that it might be due to the existence of sickle cell genes which "differ in the proportion of the abnormal hemoglobin for which they are characteristically responsible".

Other investigators have utilized other methods for studying the hemoglobin in sickle cell anemia. Singer, Chernoff and Singer (388) employed the alkali denaturation method. They found that fetal hemoglobin, which is alkali resistant, may be demonstrable in normal individuals until the end of the second year of life. In older individuals this alkali resistant hemoglobin was found regularly in those with sickle cell anemia but was lacking in all 60 instances of sickle cell trait and the normal persons who were studied. The alkali resistant hemoglobin was found also in some patients with Mediterranean anemia, hereditary spherocytosis, chronic aregenerative anemia, pernicious anemia and leukemia. The authors concluded that 4 of 11 patients with sickle cell anemia had fetal hemoglobin, while the others had a fetal-like hemoglobin They felt that the evidence warranted the recognition of three types of hemoglobin, normal (N), a type that is electrophoretically abnormal (S), and a type that is resistant to denaturation in an alkaline medium, fetal hemoglobin (F). Since their figures for the amount of fetal hemoglobin (F) in the erythrocytes of patients with sickle cell anemia agreed with the values of the normal hemoglobin (N) found by electrophoretic methods in some patients with sickle cell anemia, 5 to 20 per cent (456), it was assumed that "anemia" cells contain a mixture of "S" and "F" hemoglobin, while the "trait" cells *contain* a mixture of "N" and "S" hemoglobin (388) It was suggested that the alkali resistant hemoglobin fractions in the various disorders represented a continuation or reactivation of the production of embryonic pigment This hypothesis appears to be at variance with Watson's observation (451) that, in the newly born, few erythrocytes sickle whereas the number increases as the adult hemoglobin replaces the embryonal As possible explanations of this discrepancy, Singer and co-workers suggested that (1) the fetal hemoglobin is unable to sickle, or (2) the erythrocytes in older children contain either adult or fetal hemoglobin, but not a mixture of both *This explanation is consistent with their concept that the erythrocyte* population in sickle cell anemia is not homogenous since in survival studies some of the erythrocytes appeared to have a longer life span than others in the same patient (390)

A report by Schneider and Levin (362) supports the concept that sickle cell anemia hemoglobin differs qualitatively from both normal and sickle

cell trait hemoglobin. They reported that when erythrocytes from 2 patients with sickle cell anemia were injected into rabbits, there resulted antisera which agglutinated erythrocytes of 19 patients with sickle cell anemia, but not erythrocytes of 21 persons with sickle cell trait nor those of 124 normals. Furthermore, by use of the "direct" Coombs test, they found agglutinin antibody in the sera of all 13 patients with sickle cell anemia, and in 4 of 11 patients with sickle cell trait. It was suggested that the excessive hemolysis in the disease might be related to the presence of an agglutinin. Ingbar and Kass (208) have studied the problem in a different way and have reported a significantly greater content of reduced glutathione in the erythrocytes of patients with sickle cell anemia. They noted that prolonged saline dialysis, which removed the reduced glutathione, rendered them incapable of sickling. Sulfhydryl inhibitors inhibited sickling in vitro Therefore, they suggested that the genetically determined abnormality is related to the sulfhydryl activity within the erythrocytes.

Ponder (328, 329) has given a detailed description and discussion of the changes that occur and the forces that are involved in the sickling phenomenon. Ponder (329), Granick (161), and Perutz and Mitchison (323) have suggested that the sickled form is produced by the crystallization of reduced hemoglobin within the erythrocytes. Isaacs (209) has reported that normal erythrocytes assume the sickle form if placed in glue, but the relationship of this phenomenon to sickle cell disease is obscure at present

In summary, the sickling characteristic of the erythrocytes is related to the state of its hemoglobin since the cells assume the sickle shape under conditions of lower oxygen tension and the discoid form in the presence of adequate oxygen Furthermore, investigators who have employed different techniques such as electrophoresis, alkali denaturation, hemoglobin solubility, antiserum production, and chemical analysis, have reported differences between sickle cell anemia hemoglobin and that found in normal cells. Differences between the hemoglobin obtained from erythrocytes of sickle cell anemia and those of sickle cell trait have been noted also

Anemia

The hemolytic nature of the anemia is well established The increased fecal urobilinogen and elevated serum bilirubin observed during life and the evidence of increased blood destruction found at necropsy are difficult to explain in any other way The evidences of increased blood formation that occur in hemolytic syndromes are also found The reticulocytes and leukocytes are increased, the marrow is hyperactive, and normoblasts are often found in the circulation

At first, the anemia was thought to be due to phagocytosis of the sickled erythrocytes observed in vitro and in smears made from peripheral blood

(417, 418, 172). Since splenectomy appeared to benefit some patients, the spleen was considered to play an important role in hemolysis although it was recognized that the spleen was not the cause of sickle cell formation (172). Further experience with longer follow-up studies on patients who had splenectomy, and observations of patients who had no benefit from splenectomy, made the role of the spleen appear unimportant in the majority of patients with sickle cell anemia. Beyond these early speculations, little real progress in understanding the hyperhemolysis was made until recent years when survival of the erythrocytes was studied.

The experiments of Altman (12), Callender, Nickel, Moore and Powell (67, 68) and Singer, Robin, King and Jefferson (392), in which the survival times of transfused erythrocytes were studied, have shown that the increased hemolysis is due to a defect in the erythrocyte. Cells from normal persons survived for the normal time when transfused into patients with sickle cell anemia. In marked contrast, the average survival time of cells from patients with sickle cell anemia, when transfused into normal persons, was 15 to 60 days, only a fraction of the normal 120 days. Cells from individuals with the sickle cell trait survived normally.

The exact method of destruction of excessive numbers of these cells is unknown. Various explanations have been advanced Congested and dilated capillaries were noted by Diggs and Ching (124) as one of the most conspicuous features of the disease. Since they encountered difficulty when they attempted to resuspend sickle erythrocytes, they suggested that the capillary engorgement might be due to the elongation and interlocking of the cells which would be increased by anoxemia. This stasis and clumping of erythrocytes has been considered by some authors to be the main cause of increased hemolysis through disintegration of the enmeshed sickled erythrocytes (27, 28, 30) However, these same authors reported cases that exhibited similar capillary engorgement at necropsy without anemia and postulated that anemia developed only when bone marrow compensation failed. Anemia would not develop if blood production kept pace with blood destruction The stasis theory of hemolysis appears attractive but the available evidence makes it unlikely. Evidence of increased blood destruction is not found at necropsy in instances of sicklemia even though capillary engorgement may be present (124) Other evidence in this direction is the observation that *fecal urobilinogen excretion* and reticulocyte counts have been within the normal range in the patients with sickling trait that we have studied (246). Also against this theory is the failure of Reinhardt, Moore, Dubach, and Wade (340) to find any decrease in fecal urobilinogen after oxygen administration sufficient to reduce the numbers of sickled forms in both the arterial and venous blood In considering this point, Callender, Nickel, and Moore and Powell (68) suggested that the methods

used in determining the urobilinogen might not have been sensitive enough to detect slight decreases. Even stronger evidence against the erythrostasis congestion theory of hemolysis are the studies of the survival of transfused cells (67, 68, 392). In these studies, the survival time of sickle cell anemia erythrocytes was the same whether the recipient had sickle cell anemia, sickle cell trait, or no disease, very different conditions in which one would *not expect the degree of capillary stagnation to be the same. The same* survival time of donor sickle cell trait erythrocytes when transfused into normal recipients, and recipients with sickle cell anemia, is similar evidence.

Diggs and Bibb (123), Shen, Castle, and Fleming (378) and Nickel (68), have reported increased mechanical fragility of cells of patients with sickle cell disease when the cells are in the sickle form. Since the cells from patients with sickle cell anemia may assume the sickled form at oxygen tensions found in the circulation while the cells in the sickle trait do not (184a), the cells in patients with sickle cell anemia may be destroyed in the circulation because of their abnormal susceptibility to trauma

Singer, Motulsky, and Wile (390) have reported a case of aplastic crisis in sickle cell anemia that developed during the course of an atypical pneumonia This observation appears to indicate that the increase in anemia that sometimes occurs during crisis, and at other periods, may be the result of either marrow inhibition or increased hemolysis, or both, and affords the most acceptable explanation of the effect of infection on the peripheral blood picture

In summary, it appears that in some individuals bone marrow failure may be an important factor in anemia during crisis In most circumstances, the anemia is predominantly hemolytic in nature. The excessive hemolysis is a result of the defective erythrocytes in sickle cell anemia. The exact manner and site of erythrocyte disintegration is unknown The actual destruction of the cells may take place in the general circulation because of their abnormal susceptibility to trauma when in the sickled form.

Symptoms

In sickle cell anemia, symptoms arise from any organ or system in the body, sometimes quite suddenly When attempts were made to explain the clinical manifestations, it soon became apparent that they could not be explained by the anemia, and more satisfactory explanations were sought.

As Diggs and Ching (124) have indicated, the lesions found at necropsy are of two general types One group is related to hemolysis and the other

hyperplasia of the blood forming organs. The lesions due to circulatory

vascular changes are also quite prominent. Capillary engorgement in the liver, spleen, kidney, lungs and other organs suggests that packing of the distorted erythrocytes has occurred under pressure (124). Infarcts are often present in the spleen, brain, lung and kidney (124). At times thrombi have been demonstrated in the lung, brain, and other organs (117, 126, 136, 142, 153, 124, 200, 273, 406, 476). However, other cases have been reported where areas of infarction occurred in the absence of demonstrable thrombosis (28, 42, 81, 117, 13) Kimmelstiel (229) has made a critical review of the reports on this aspect of the disease, and concluded that actual thrombosis is rare while ischemic necrosis without thrombosis is common Others have reached similar conclusions (426, 334).

The widespread capillary congestion associated with areas of necrosis and hemorrhage has suggested that the clinical evidence of involvement of the various systems in the body might be explained by these vascular lesions. The abnormal configuration of the erythrocytes, which would probably be most marked in organs where blood flow and oxygen tension are reduced, has been thought to cause interlocking and entrapment of the erythrocytes in the narrow capillaries that become distended in the process (28). Others have placed a different interpretation on these observations The dilated vessels and sickled erythrocytes have been considered to be secondary to shock in which vascular spasm produced a sequence of stasis, small vessel dilatation, conglutination of the erythrocytes, anoxia, and sickling (426)

While the exact sequence of events remains in doubt, at the present time it seems that infarction with necrosis and hemorrhage is the best explanation for the widespread involvement of the various systems that occurs in this disease. The clinical picture depends on the organs involved and the severity and duration of the anemia.

CLINICAL MANIFESTATIONS

General

Sickle cell disease is hereditary. As a rule, inheritance of the gene in the heterozygous state results in the generally benign sickling trait while inheritance of the gene in the homozygous state produces the disease, sickle cell anemia Recent studies of various kinds have indicated that the fundamental defect is an abnormal hemoglobin which in its reduced form assumes a peculiar sickled shape and distorts the envelope of the erythrocyte If it is sufficiently marked, this intrinsic defect of the erythrocyte leads to its premature destruction in vivo by some subtle mechanism not yet established with certainty As a result of this process the patient develops the characteristic features of a hemolytic process such as anemia, hyperbili-

used in determining the urobilinogen might not have been sensitive enough to detect slight decreases Even stronger evidence against the erythrostasis congestion theory of hemolysis are the studies of the survival of transfused cells (67, 68, 392). In these studies, the survival time of sickle cell anemia erythrocytes was the same whether the recipient had sickle cell anemia, sickle cell trait, or no disease, very different conditions in which one would not expect the degree of capillary stagnation to be the same. The same survival time of donor sickle cell trait erythrocytes when transfused into normal recipients, and recipients with sickle cell anemia, is similar evidence.

Diggs and Bibb (123), Shen, Castle, and Fleming (378) and Nickel (68), have reported increased mechanical fragility of cells of patients with sickle cell disease when the cells are in the sickle form. Since the cells from patients with sickle cell anemia may assume the sickled form at oxygen tensions found in the circulation while the cells in the sickle trait do not (184a), the cells in patients with sickle cell anemia may be destroyed in the circulation because of their abnormal susceptibility to trauma

Singer, Motulsky, and Wile (390) have reported a case of aplastic crisis in sickle cell anemia that developed during the course of an atypical pneumonia This observation appears to indicate that the increase in anemia that sometimes occurs during crisis, and at other periods, may be the result of either marrow inhibition or increased hemolysis, or both, and affords the most acceptable explanation of the effect of infection on the peripheral blood picture

In summary, it appears that in some individuals bone marrow failure may be an important factor in anemia during crisis. In most circumstances, the anemia is predominantly hemolytic in nature. The excessive hemolysis is a result of the defective erythrocytes in sickle cell anemia The exact manner and site of erythrocyte disintegration is unknown. The actual destruction of the cells may take place in the general circulation because of their abnormal susceptibility to trauma when in the sickled form

Symptoms

In sickle cell anemia, symptoms arise from any organ or system in the body, sometimes quite suddenly. When attempts were made to explain the clinical manifestations, it soon became apparent that they could not be explained by the anemia, and more satisfactory explanations were sought

As Diggs and Ching (124) have indicated, the lesions found at necropsy are of two general types One group is related to hemolysis and the other to circulatory changes Included in the hemolytic group are the hemosiderosis of the liver, cholelithiasis, siderofibrosis of the spleen, increased phagocytosis in the reticuloendothelial system and the changes associated with hyperplasia of the blood forming organs. The lesions due to circulatory or

facial features, and subnormal head circumference are additional characteristics. One individual in their series who was thought to have sickle cell trait manifested similar findings. Winsor and Burch (465) studied 15 patients with sickle cell anemia between the ages of 6 and 32 years and four patients between the ages of 30 and 47 years with sicklemia. They utilized the clinical picture, photographs, roentgenograms and anthropometric measurements. In summarizing their findings, they stated: "The adults have linear builds, emaciation, long legs, 'hoop chest', short trunks, 'spider hands', lumbar lordosis and upper dorsal kyphosis, tendencies to hypogonadism and general appearance of fragility. The children have 'hoop chest', enlarged protruding abdomens and thin legs. Changes encountered in adults may be present in children if the disease is severe." The habitus of patients with sickle cell anemia was considered to be secondary to the disease rather than primary. The scaphocephalic (182), dolichocephalic (349) and macrocephalic (165) skulls that have been described were not found to be typical of sickle cell anemia.

Apparently, the body habitus depends to some extent on the severity and duration of the disease for some adults with the disease, particularly the milder forms, have normal physical and mental development. The fertility of women with the disease has been thought by some observers to be lower than normal (112, 401). The modern techniques of endocrine evaluation such as 17-ketosteroid excretion, gonadotrophin excretion, etc., might help clarify the relationships between body build, sexual maturity, endocrine function and sickle cell disease.

Skin

The most common cutaneous lesion in sickle cell anemia is ulceration of the lower extremities. Ulcers were noted in all of the first four case reports (195, 449, 94, 278). The subject has been discussed in detail by some authors (109, 264) and has been the subject of case reports by others (49, 107, 201, 230, 238, 364, 365, 407). With the exception of the scars of previous ulcers, other dermatological lesions are rare. Multiple, tender, subcutaneous swellings have been described on the extremities and forehead, and areas of hyperpigmentation and vitiligo have been observed (264).

Although one report mentions an instance of ulceration during the first year of life (264), ulcers are rare in young children and most often develop in the second decade (109, 264). It has been estimated that active lesions are present in 25 per cent of patients with the disease (264), while 75 per cent of adults have either ulcers or residual scars (124). Ulceration has been noted in Caucasians who had the disease (475). The incidence of cutaneous ulceration in Negroes with the sickle cell trait appears to be the same as that in normal Negroes (122).

Most often the ulcerated area is a single lesion on the medial aspect of

rubinemia, increased fecal urobilinogen, together with evidence of efforts at compensation—marrow hyperplasia and reticulocytosis.

This series of events is lifelong for the unfortunate victim, but both the clinical manifestations and the anemia vary among patients. Some succumb to the disease in childhood. Others survive adolescence although they remain handicapped by a retarded development and mild chronic illness. A few lead relatively normal lives but even these rarely survive beyond middle age. The reason for this difference in severity of the disease in different patients is unknown. It may be due to quantitative differences in the various types of hemoglobin.

In the majority of patients, both children and adults, the state of mild chronic illness is interrupted by *episodes of more severe symptoms*, the periods of "crisis" They may be mild, last only a few days, and consist of nothing more than fever and mild abdominal pain or arthralgia. When the episode is more prolonged, rheumatic fever may be simulated. At other times, the patient suddenly becomes ill with severe abdominal pain, nausea, vomiting and fever that may suggest an acute surgical emergency Hemiplegia, convulsions, or coma may focus attention on a catastrophe in the central nervous system The factors which initiate these dissimilar episodes remain unknown for the most part, as do the reasons for involvement of the various systems at any particular time During these periods of "crisis", sometimes called "hemolytic crisis", the anemia and jaundice may increase rapidly. At other times, even when clinical manifestations of crisis are present, the blood counts and serum bilirubin may remain unchanged *If they become altered during such an episode, they usually return to* their previous levels after recovery from the "crisis". In any individual, the degree of anemia tends to remain stable over a period of years, perhaps for life

Up to the present time, the various forms of treatment have been able to do no more than temporarily alter the course of the disease which sooner or later ends in death The childhood deaths usually occur during an episode of "crisis" or in the course of some infection. Heart failure, pregnancy, vascular accidents and uremia are added hazards for adults with the disease

Development

As early in 1922, the association of a slender habitus and scant axillary and pubic hair was noted in a case of sickle cell anemia (278). Since then, other evidences of retarded development have been observed. Sharp and Vonder Heide (365) called attention to the eunuchoid habitus in a group of 32 patients, 30 of whom had sickle cell anemia They noted "long, tapering fingers, slender feet, sparse hair growth except on the head, and marked sexual underdevelopment for the age" Subnormal weight, *small*

pitched, and transmitted to the left axilla. Systolic murmurs in the aortic and pulmonary areas are next in frequency (464). Although they are relatively rare, diastolic murmurs have been noted in the mitral area (231, 233, 248, 464), and less often in the aortic and pulmonic regions (464). An explanation of the murmurs has not been given by post mortem examinations, which have failed to reveal any endocardial lesions in the uncomplicated cases (157, 179, 233, 303, 426, 474). Lesions on the corpora arantii and mitral valve have been found in the presence of co-existing diseases (164, 447)

Cardiac enlargement has been detected clinically in 40 per cent (15, 16), 59 per cent (197) and 91 per cent (464) of patients with the disease. At times the enlargement is evident on physical examination, but more often it is apparent only on roentgenological examination, which revealed cardiac enlargement in 95 per cent of one series (464). In a group of 36 patients, mentioned in two reports (233, 464), enlargement of both the right and left ventricle was reported in over 85 per cent, while enlargement of the pulmonary conus occurred in about 73 per cent. Enlargement of the left auricle has been reported in association with rheumatic fever (149, 173) and as a rare occurrence in uncomplicated sickle cell anemia (166) Necropsy nearly always reveals cardiac enlargement of some type, rarely associated with any lesion of valves, endocardium, or pericardium (104, 124, 179, 189, 233, 279, 426, 464, 474, 477). The enlargement is due mainly to dilatation, but heart weights as high as 480 grams have been found (233)

The changes in the electrocardiogram in sickle cell anemia are non-specific in nature. The rate is frequently increased and the rhythm is usually regular. Occasionally, auricular premature beats occur and auricular fibrillation has been reported (190). Nodal rhythm with a ventricular rate of 140 occurred in one patient we observed The most frequent abnormality is prolongation of the PR interval reported in 12 per cent (464), 25 per cent (166), and 50 per cent (233) of three series of cases Moderate prolongation is not uncommon and intervals up to 0 24 have been reported (81, 231, 233, 464, 480) No reports of complete AV block or bundle branch block have been found. In one series of 25 patients (464) no displacement of the ST segment of as much as 1 mm was observed, while in another series of 30 patients (190) displacement of the ST segment and abnormalities of the T waves were considered to indicate myocardial ischemia in 12 patients. Inversion of the T wave in leads I and II is rare but low amplitude of the T waves (0 2 mm) in leads I and II has been noted, as has inversion of the T wave in lead III and in the precordial leads (104, 401) Chronic anoxia with an increase in vagal tone has been suggested as a cause of the delayed auriculo-ventricular conduction time in these patients (233) The return of inverted T waves to normal following repeated transfusions has been reported (190)

the lower leg near the ankle. Not infrequently, the ulcers are bilateral or multiple, and any portion of the leg, the dorsum of the foot, or the elbows may be involved. They vary in diameter from one to many centimeters, and the outline may be round, oval or irregular. The margins are usually sharply defined, often slightly elevated and may be surrounded by induration. The ulcers are usually described as "punched-out" in appearance, but the base of granulation tissue may project above the level of the surrounding skin. The base of the ulcer usually is grayish from the exudate that covers the granulation tissue. The ulcer is chronic. Periods of several months or a year may elapse before healing occurs. The residual scar is shiny, atrophic and depigmented. There may be hyperpigmentation in the surrounding tissue.

The factors responsible for the ulcers and their chronic course are somewhat obscure. The location of the lesions and the frequent history of antecedent trauma suggests that such an event may be a factor in initiating the lesion. Capillary blockage and vascular thrombosis have been considered the main factors responsible for the ulceration (264). However, as many authors have pointed out, similar lesions occur in familial spherocytic anemia where no unusual tendency to thrombosis or capillary engorgement exists. Histologic study of biopsy material (201, 415, 109, 124) has revealed no unusual vascular lesions. The picture has been reported as a non-specific one of chronic granulomatous inflammation of the skin with ulceration. Cummer and LaRocca have reported the histological appearance in detail (109) The importance of the anemia per se as a factor in ulceration is difficult to evaluate since in few other conditions does anemia begin so early in life and pursue such a chronic course.

Heart

A systolic murmur and cardiac hypertrophy manifested by an "apical impulse in the sixth interspace one inch outside the midclavicular line" were reported in the original paper on sickle cell anemia (195). Cardiac enlargement was found in the first patient that came to necropsy (417). This aspect of sickle cell anemia has been noted in subsequent case reports (179, 476, 480, 149, 164, 189, 157, 217, 201, 231) and has formed a part of reports of larger series of patients (166, 190, 197) Several authors have presented detailed discussion of the cardiovascular system in sickle cell anemia (196, 233, 464, 469)

The most common cardiac abnormality in sickle cell anemia is a murmur (17, 149, 228, 231, 248, 417, 464) which has a reported frequency of as high as 95 per cent (196) and 100 per cent (233) The commonest murmur is an apical one that occurs during systole (233, 464) In some instances there is only a soft, localized murmur, but in others the murmur is loud, high-

pitched, and transmitted to the left axilla. Systolic murmurs in the aortic and pulmonary areas are next in frequency (464). Although they are relatively rare, diastolic murmurs have been noted in the mitral area (231, 233, 248, 464), and less often in the aortic and pulmonic regions (464). An explanation of the murmurs has not been given by post mortem examinations, which have failed to reveal any endocardial lesions in the uncomplicated cases (157, 179, 233, 303, 426, 474). Lesions on the corpora arantii and mitral valve have been found in the presence of co-existing diseases (164, 447).

Cardiac enlargement has been detected clinically in 49 per cent (15, 16), 59 per cent (197) and 91 per cent (464) of patients with the disease At times the enlargement is evident on physical examination, but more often it is apparent only on roentgenological examination, which revealed cardiac enlargement in 95 per cent of one series (464). In a group of 36 patients, mentioned in two reports (233, 464), enlargement of both the right and left ventricle was reported in over 85 per cent, while enlargement of the pulmonary conus occurred in about 73 per cent Enlargement of the left auricle has been reported in association with rheumatic fever (149, 173) and as a rare occurrence in uncomplicated sickle cell anemia (166). Necropsy nearly always reveals cardiac enlargement of some type, rarely associated with any lesion of valves, endocardium, or pericardium (104, 124, 179, 189, 233, 279, 426, 464, 474, 477) The enlargement is due mainly to dilatation, but heart weights as high as 480 grams have been found (233)

The changes in the electrocardiogram in sickle cell anemia are non-specific in nature. The rate is frequently increased and the rhythm is usually regular Occasionally, auricular premature beats occur and auricular fibrillation has been reported (190). Nodal rhythm with a ventricular rate of 140 occurred in one patient we observed. The most frequent abnormality is prolongation of the PR interval reported in 12 per cent (464), 25 per cent (166), and 50 per cent (233) of three series of cases. Moderate prolongation is not uncommon and intervals up to 0 24 have been reported (81, 231, 233, 464, 480). No reports of complete AV block or bundle branch block have been found. In one series of 25 patients (464) no displacement of the ST segment of as much as 1 mm was observed, while in another series of 30 patients (190) displacement of the ST segment and abnormalities of the T waves were considered to indicate myocardial ischemia in 12 patients Inversion of the T wave in leads I and II is rare but low amplitude of the T waves (0 2 mm) in leads I and II has been noted, as has inversion of the T wave in lead III and in the precordial leads (104, 401) Chronic anoxia with an increase in vagal tone has been suggested as a cause of the delayed auriculo-ventricular conduction time in these patients (233) The return of inverted T waves to normal following repeated transfusions has been reported (190)

Despite the frequency of right ventricular enlargement, right axis deviation occurs rarely (464). The absence of right axis deviation and the lack of significant changes in the P waves may be minor but helpful points in distinguishing sickle cell anemia from rheumatic fever with mitral stenosis.

The cause of the cardiac abnormalities in this disease is an interesting problem. The autopsy findings, such as vascular changes in the lung (476), fundi (182), obliterative endarteritis of the coronary arteries (464), minor valvular abnormalities (406, 447, 164, 464), and the generalized engorgement and tortuosity of the capillaries (124), seem inadequate to explain the hypertrophy and dilatation that usually involve both sides of the heart (464), since these lesions are not observed in every case of cardiac enlargement. It is difficult to evaluate the importance of the commoner autopsy findings, such as fragmentation of the myocardial fibers (426, 476), fatty degeneration and scarring (129, 279, 426, 474), cellular infiltration with necrosis (406). As has been emphasized by Klinefelter (233) and Wintrobe (469) cardiac enlargement, systolic and diastolic murmurs, congestive heart failure, electrocardiographic changes, and angina pectoris have been noted in other types of anemia, such as that due to chronic blood loss, pernicious anemia, and chronic hemolytic jaundice. Ludke and Schuller (257) have reported the development of cardiac hypertrophy secondary to anemia produced experimentally in dogs. Severe anemia may weaken the heart muscle and at the same time increase the work of the heart because of the increased cardiac output (53). In sickle cell disease the anemia is of longer duration and is more resistant to treatment than is the case with most other forms of anemia Perhaps for this reason the changes in the heart occur more frequently In sickle cell anemia, cardiac complications are most common in the patients who have severe grades of anemia of long duration It appears that, while other lesions may be responsible for the cardiac manifestations in an occasional patient, chronic anemia is the most acceptable explanation for the majority of the cardiac abnormalities in sickle cell anemia

At times the manifestations of congestive failure may dominate the picture in a patient with sickle cell anemia In other patients, rheumatic carditis may be simulated and this led to a diagnosis of rheumatic fever in 39 per cent of one series (197) The co-existence of rheumatic fever and sickle cell anemia has been reported four times (149, 405, 406, 447) and the differential diagnosis has been discussed (173). With the many reports of the frequency of infarcts in various organs and the tendency toward thrombosis, it is interesting that angina pectoris and coronary thrombosis evidently are rare in this disease. There has been one report of a patient with a picture that suggested coronary thrombosis but this was not proved (480). Angina pectoris appears to be very rare, perhaps because the anemia

in this disease occurs for the most part in younger persons who do not have much coronary sclerosis

Gastro-intestinal System

Symptoms and signs referable to the gastro-intestinal system occur at some time during the course of the disease in nearly every patient with sickle cell anemia Often a history of jaundice, anorexia, failure to gain weight, abdominal pain, nausea, vomiting and constipation may be elicited. In one group of 92 patients (197), 35 per cent complained of some disturbance of the gastro-intestinal tract The complaints may be so misleading that the initial diagnosis may be erroneous. The initial incorrect diagnoses in one series (197) were acute appendicitis, perforated viscus, intestinal obstruction, acute cholecystitis, cholelithiasis, catarrhal jaundice, acute hepatitis, acute gastro-enteritis, intestinal parasites and bacillary dysentery.

Abdominal pain has been reported to have an incidence of 50 per cent (15, 16) and 52 per cent (166). It has been listed as a frequent finding by a number of authors (15, 16, 17, 101, 190, 195, 197, 248, 256, 278, 292). Abdominal pain noted in sickle cell anemia tends to be recurrent It may be sudden in onset and so severe that the administration of spinal anesthesia is necessary to give relief when narcotics fail to do so The pain is frequently sharp, intermittent and moderately severe, at other times it is constant Although it is more frequent and more severe during crisis, it also occurs when other manifestations of an acute exacerbation are absent. The location of the pain is variable It may be in any part of the abdomen Epigastric pain has been noted specifically following the ingestion of food (248). Left hypochondrial pain is frequently observed and pain localized in the right lower quadrant or right upper quadrant is not unusual Vomiting, fever, leukocytosis, icterus with localized tenderness and spasm are often also present and suggest the need for surgical intervention.

Various explanations have been offered for the abdominal pain. Probably the cause of the abdominal pain is not the same in every patient. Pain, particularly over the left hypochrondrium, has been explained on the basis of splenic hemorrhage which has been demonstrated at autopsy (418) All abdominal pains cannot be explained on this basis since splenectomy does not eliminate this type of pain in all cases (248). Hepatic arterial thrombosis has been considered as the explanation in one case with pain in the liver area (477), but it would appear that this is quite rare. Gall stones occur frequently in patients with sickle cell disease (69, 189, 257, 190, 197, 453) and may be responsible for the attacks of abdominal pain in certain instances They do not occur in all patients with sickle cell disease and thus do not afford an adequate explanation for the abdominal pain in all cases.

Bone and Joint Manifestations

Pains in the joints or extremities are among the commoner clinical manifestations of the disease. In one review (264) "joint or myalgic pains" were present in 186 of 214 subjects. In another study of 48 patients, who had 128 admissions to the hospital, "joint or extremity pain" was the chief complaint on 32 admissions and an associated, prominent symptom on 17 others (166). Other reports have given an incidence of skeletal manifestations of 51 per cent (16, 17) and 55 per cent (15).

Although pain in the extremities may be severe enough to incapacitate the patient temporarily, more often it is aching in character and not so severe. It is usually in the muscles and the long bones rather than in the joints (197). In interesting contrast to the frequency of symptoms referable to the extremities is the fact that objective joint manifestations are seldom mentioned in the literature. Actual tenderness, heat and swelling are unusual

The symptoms from bones and joints may lead to an erroneous diagnosis It has long been recognized that sickle cell anemia can produce a picture that may be indistinguishable from rheumatic fever. Also, there has been a report of a patient who appeared to have moderately severe rheumatoid arthritis with fusiform swelling of the fingers, atrophy, contractures and roentgenographic changes (93) We have observed a similar patient. Whether such cases represent the coincidental occurrence of two diseases or changes due to sickle cell anemia alone is not clear. The fever, leukocytosis, and tenderness over an extremity have lead to a diagnosis of acute osteomyelitis (316, 258) The discussion of the skeletal lesions is included in the discussion of the "Roentgenology"

Genito-urinary System

Although symptoms referable to the genito-urinary system are not common in sickle cell anemia, those that do occur are often dramatic and serious. One of these is spontaneous hematuria which has attracted attention only in recent years during which time 13 patients have been reported (3, 28, 155, 190, 316, 354, 462). Goodwin, Alston and Semans, who observed four patients and found hospital records of three others, have given the most complete discussion of the subject (155) Only cases with macroscopic [illegible] patients [illegible] istances, the bleeding has been severe, unilateral, and recurrent, sometimes returning over a period of years In some patients, the pyelograms were normal, but *in others* they suggested a tumor or blood clot in the renal pelvis Operation for suspected tumor has been performed twice (3, 155). The cause of the bleeding is not clear. Infarction has been suggested (462),

but the microscopic studies of the kidney in three subjects (3, 155) have not revealed infarction. The sections revealed dilated, congested vessels with slight erosion of the mucosa of the papillae. In one patient, the vascular congestion and packing was thought to be responsible for massive cortical necrosis (28). It is interesting that four of the patients may well have had sickle cell trait rather than sickle cell anemia (3, 155, 354). Hematuria is one of the rarer manifestations of sickle cell disease In one series of 54 patients, hematuria was listed as occurring three times (190), while in other series of 92 patients (197), 48 patients (166) and another of 214 collected from the literature (264), hematuria was not mentioned.

In 1934, Diggs and Ching (124) mentioned the necropsy findings in a patient with sickle cell anemia who suffered from priapism. Since that time, this has been noted in 23 patients with sickle cell anemia, and the disease has been recognized as one of the possibilities when the symptom is present (69, 115, 154, 166, 185, 190, 197, 227, 249, 308, 316, 324, 352). The youngest patient reported was 8 years of age (115). The duration of the painful erection has generally been from 10 to 36 days. In at least 5 patients, complete impotence followed (185, 227, 249, 308, 352). In all probability follow-up studies would reveal that the residual impotence is much commoner since priapism of over two days' duration usually, but not invariably, leads to this result (227). It is interesting that painful erections of shorter duration preceded the episode of priapism in many of the reported cases, and at least one patient retained some degree of potency following treatment for priapism by aspiration of the penis (185) The pathogenesis of the priapism has been discussed by several authors (154, 227), and it has been suggested that circulatory stasis and vascular engorgement lead to thrombosis and the eventual fibrosis reported at necropsy (124) We have found no reports of priapism in persons with sickle cell trait.

Other renal abnormalities are more common but less dramatic Slight albuminuria and a low fixed specific gravity have been noted (190, 197) Despite the frequent fixation of the specific gravity in both children and adults (190), marked nitrogen retention rarely occurs except when severe liver damage is also present (190). We have seen two patients, adult women, with marked nitrogen retention, one of whom came to autopsy. In both instances a *coincidental severe pyelonephritis was the most logical explanation* of the renal insufficiency. Degenerative and proliferative changes have been reported at autopsy (124, 476), but there have been few studies of renal function. Calcagno, McLany and Kelley (65) studied the glomerular filtration rate by means of the sodium thiosulfate method In all five subjects with sickle cell anemia, aged 4 to 15, in whom the hemoglobin ranged from 6.5 to 7 2 grams, the glomerular filtration was found to be from 30 to 60 per cent below normal. Normal values were found in two children with

the sickle cell trait and an adult who had a comparable degree of anemia secondary to uncinariasis. Henderson (190) found normal urea clearances in two patients. Normal phenolsulfonphthalein excretion was noted in one patient and retention of the dye was found in another who was recovering from crisis Further studies of renal function are needed. The influence of repeated episodes of crisis on the function of the renal tubules cannot be assessed at present.

The clinical significance of the post-mortem findings is unknown. Marked congestion of the capillaries and arterioles of the glomeruli and tubules is an almost constant finding Different authors have reported the occurrence of glomerulonephritis (233), a peculiar type of nephritis (233), acute glomerulitis (30), sclerosis of glomeruli (157, 576) and areas of hemorrhage involving the glomeruli (157) In the epithelial cells of the convoluted tubules, in the cells of Henley's loop, and in the collecting tubules, granules of amorphous, yellow-brown pigment have been observed (124, 233, 279, 302) The pigment gives a variable reaction to iron (124, 233, 279). In various patients the tubules have shown degeneration (474), necrosis (30), atrophy (157), and calcification of the degenerating epithelium (426, 476). Areas of ischemic necrosis in the cortex have been reported (229). Diggs and Ching (124) state that the degenerative and proliferative changes in the kidney are absent or minimal in the early or mild cases, but are so constantly found in the cases of long standing that they form a part of the picture

Central Nervous System

The first instance of central nervous system involvement in sickle cell anemia was reported by Sydenstricker, Mulherin and Houseal in 1923 (418) In 1940, Hughes, Diggs and Gillespie (103) reviewed the literature on this aspect of the disease. Their report included six patients that they had observed and 25 reported previously (15, 19, 20, 23, 46, 37, 95, 104, 150, 157, 177, 222, 223, 248, 320, 321, 355, 393, 418, 476) We have found reports of 14 additional patients since that time (92, 156, 203, 354, 356, 424, 446, 447, 451, 478) In all probability, central nervous system complications are more common than these reports imply We have observed severe involvement of this type in three of a group of 50 patients seen in the last 10 years. In other series it has been mentioned as occurring in 2 of 30 patients (215), in one of 54 (190), in 18 of 92 (197), and thirteen times in 129 admissions of 48 patients (166) In one study of the brains of five unselected cases of sickle cell anemia, lesions were found in the central nervous system in each (457)

The age of the reported patients with central nervous system involvement varied from 18 months (37, 150) to 62 years (55). In at least two

instances, symptoms have occurred in patients with sicklemia who had no anemia at the time (354, 424).

In their review of 31 patients, Hughes, Diggs and Gillespie (203) listed 29 symptoms. Drowsiness, stupor or coma occurred in about one-half of the patients, as did hemiplegia. Aphasia, headache, convulsions, and stiffness of the neck were each found in about one-fourth. Pains in the extremities, back and neck were common. Nystagmus, pupillary changes, ptosis, dysphagia and generalized rigidity were each noted several times. Diplopia, hemianopsia, cranial nerve paralysis and delirium occurred once. Mental deficiency and psychotic tendencies have been observed (46, 55, 166) The neurological disorder may subside completely or may be the cause of death during the episode. Spastic paralysis may be the result in those who recover. In many, there have been recurrent episodes, sometimes of increasing severity.

The cerebrospinal fluid may be normal during a severe attack of this type, or increased pressure, bloody fluid, xanthochromia, sickled erythrocytes, increased protein or increased leukocytes may be present (103).

The electroencephalographic findings in 38 patients with sickle cell anemia have been reported (198). The EEG's were considered to be abnormal in 26 Abnormal tracings were found in all children under six years of age. Only three patients over 14 years of age were studied and all had normal EEG's. There were 9 patients with a past history of nervous system symptoms who had normal tracings. The abnormal tracings were thought to be due primarily to cerebral anoxia and secondarily to degenerative processes in the nerve cells.

Various explanations have been offered for the lesions in the brain and spinal cord. Endarterial proliferation (19, 55), thrombosis of the larger vessels (55, 96, 424), massive hemorrhage (354, 447), cortical atrophy (92, 203), fat embolism (442, 446, 457) and areas of hemorrhage and necrosis (55, 229, 424) have been observed at necropsy. On the basis of histological study, vascular congestion, stasis, thrombosis, infarction, and atrophy have constituted the sequence of events accepted most often (19, 92, 203, 222, 223) Others interpret the findings as indicating vasospasm and ischemic necrosis (229). The endothelial proliferation in the arteries has been considered the primary factor (55), but this vascular change is reported by other authors in only a minority of patients (229). At times, necropsy has revealed no explanation for the focal areas of tissue loss and massive hemorrhage (447). The lesions found in the central nervous system at autopsy have been discussed by Wertham, Mitchell and Angrist (457).

The involvement of the central nervous system may produce the initial manifestations in patients with sickle cell anemia and the underlying disease may not be recognized Encephalitis, meningitis, poliomyelitis, idio-

pathic epilepsy, hypoglycemia, arteriosclerosis, cerebral embolism, psychosis, syphilis, and brain tumor have been mistaken diagnoses (197, 156, 223, 229, 264, 457).

Spleen and Lymph Nodes

In sickle cell anemia the spleen exhibits a variety of changes that are unusual for an organ in a single disease. The size varies from marked enlargement to extreme atrophy (39, 92, 104, 124, 229). A spleen that weighed 1850 grams has been removed from a 10 year old child (380). At the other extreme, no spleen could be demonstrated in a 51-year-old man at autopsy (124), and in a 23-year-old woman, the spleen weighed 0.87 gram (104). In different series, the incidence of enlargement of the spleen has been reported variously as 6 per cent (166), 21 per cent (197), 30 per cent (17), and 40 per cent (166). Since splenomegaly occurs much more often in children than in adults, figures as to its incidence have little meaning unless the ages of the patients are taken into consideration. Although it has been stated that the spleen is usually enlarged in patients under 4 years of age and is seldom palpable in patients older than 7 years (17), factors other than age are important in determining the size of the spleen since fibrotic atrophy may occur in childhood (124) and splenomegaly may be found in the fourth decade (426) In some patients, the spleen has been palpable during an episode of crisis, and not palpable after the acute symptoms have subsided (17, 34, 166) Diggs and Ching (124), Diggs (121) and Mathews (279) have given detailed descriptions of the changes that occur in the spleen.

Since hemorrhage of the spleen was demonstrated in the first patient that came to necropsy (418), and later infarcts were found to be common (124), the spleen has been blamed for the abdominal pain that occurs in many patients It seems likely that the marked changes that occur in the spleen might produce symptoms in the left upper quadrant, but it is not solely responsible for symptoms in the left upper quadrant since the pain may persist after splenectomy (190)

Soon after the hemolytic nature of the anemia was recognized, the role of the spleen in the hemolysis and the possible benefits of splenectomy were considered Hahn and Gillespie (172) reported the first splenectomy and noted that improvement followed although the sickling characteristic of the cells persisted There have been reports of 24 splenectomies with variable results (37, 102, 45, 85, 91, 88, 96, 170, 171, 240, 241, 248, 339, 380, 410) With the possible exception of the patients with marked splenomegaly, it is evident that the spleen plays an insignificant role in the genesis of the anemia. In many patients the course of the disease appears unaltered by either surgical splenectomy or "auto-splenectomy". Rarely a

greatly enlarged spleen may alter the disease by aggravating the anemia and in such cases splenectomy may prove of benefit (380).

Generalized enlargement of the superficial lymph nodes, that rarely measure more than 2 cm., occurs in many children with the disease. The nodes are usually firm, discrete and non-tender. They rarely produce symptoms and appear to be of little or no importance in the disease (17). Phagocytosis of erythrocytes, hyperplasia, edema, pigmentation, fibrosis (124, 279, 406, 426), non-specific lymphadenitis (157), and hemorrhage (426) have been observed at autopsy. Enlargement of the thymus has been found several times (356, 415).

ROENTGENOLOGY

Since the first reports of abnormal roentgenograms in sickle cell anemia in 1929 by Rose (349) and Moore (288), articles on this aspect of the disease have been numerous, many based on a single patient or a small group of patients (8, 114, 179, 182, 193, 237, 454, 458). Larger series of patients have been the basis for detailed, well illustrated reports of the frequency of the various abnormalities and discussions of their pathogenesis by Diggs, Pulliam and King (126), Macht and Roman (268), Legant and Bell (247), and Carroll and Evans (82). The roentgenological aspects of sickle cell disease, thalassemia and other hemolytic disorders have been compared in several papers (165, 288, 252, 78, 444, 142). The similarity of the roentgenograms of the skull in these disorders to those made from the skulls of ancient Mayan Indians and Peruvian mummies has been noted (288, 461).

Although most of the interest in the roentgenological aspects of the disease has centered on the skeletal abnormalities, other abnormalities that are less suggestive of sickle cell anemia probably occur more often. In this category are cardiac enlargement, hepatomegaly, cholelithiasis, splenomegaly, splenic calcification, pulmonary infarction, pulmonary congestion and pneumonia. The commonest is cardiac enlargement which has been found in 50 per cent (247), 70 per cent (82), and 76 per cent (268) of several series

While the roentgenograms of the bones of a few patients with sickle cell anemia show moderate or striking changes, no abnormalities in size, shape or density can be demonstrated in the majority of patients (126, 82). No definite correlation between the severity or duration of disease and the roentgenological findings has been found, since severe anemia may be associated with no osseous abnormalities and mild anemia may be associated with advanced changes (82, 126) Demonstrable changes in the long bones have been noted in 65 per cent (268) and 55 per cent (82), in the short bones 52 per cent (268), in the spine 43 per cent (82) and 17 per cent (268). Skeletal abnormalities have been found at ages varying from

8 months (114) to 51 years (126) and 60 years (142) We have found no reports of roentgenologic abnormalities in individuals with sickle cell trait

Most authors have considered the problem of osseous lesions from the aspect of the anatomic structure involved A more fundamental approach has been offered by Macht and Roman (268), who have classified the osseous lesions demonstrable by roentgenogram into three types. The first includes those due to hyperplasia of the erythropoietic elements, the second includes those thought to be due to thrombosis, and the third, those considered to be the result of disturbances in growth.

Changes that appear to be the result of hyperplasia of the bone marrow, which is a conspicuous feature at necropsy (126), may occur in long, short, and flat bones. Widening of the medullary spaces, thinning of the cortex and irregular trabeculation have been found most often in the long bones (126, 132, 258). Marked thinning of the cortex appeared to be responsible *for a pathological fracture of the femur in one case (8). Softening of the* bony structure and irregular trabeculation have been noted in the short bones, ribs, the scapulae and pelvis (268). Of the flat bones, the skull has presented the most striking changes Increase in height and decrease in lateral measurement produce the "turricephalic" or tower skull that has been observed frequently Widening of the diploe, thickening of the parietal and frontal bones and thinning of the outer table occur most often (126, 268). The well known "hair on end" appearance of the increased radial striations is unusual (126, 268) Generalized osteoporosis of the spine may lead to vertebral flattening or cupping and kyphosis (126, 247, 258) Collapse of vertebrae has been reported (193) In discussing this development, Henken (193) suggested that the very cellular marrow is the site of vascular congestion, thrombosis and infarction which results in the formation of connective tissue replacement and weakened bone Such bone, when subjected to excessive stress, may show a tendency to new bone formation Thus, bone resorption, manifested by a decrease in the number of bone trabeculae and bone density, may be accompanied by new bone formation which increases the size of the remaining trabeculae and coarsens trabecular structure He considered the picture to be one of osteoporosis with insufficient osseous matrix

Other changes, observed in the long and short bones, have been interpreted as the result of thrombosis and infarction (247, 268) These include cortical thickening, narrowing of the medullary cavity, loss of bone tissue, periosteal reaction, and necrosis and absorption of bone. Well defined areas of rarefaction in the skull that resemble multiple myeloma or metastatic malignancy have been reported (126) Cyst-like areas have been reported in the tubular bones of the hand (454) Areas of bone destruction in the metatarsals have been reported (114, 268), and we have seen lesions in

the hand. Clinical symptoms may precede the roentgenologic changes of this type by three weeks (268). Normal roentgenograms were obtained after 6 weeks in one patient (114), and only after 8 months in another (268). Similar areas of rarefaction in a vertebra did not become apparent until 8 weeks after a crisis associated with abdominal pain, and did not heal completely until 18 months had elapsed (247). Destruction of a joint may occur (82, 247, 268) and early Charcot joints have been diagnosed infarction (247, 268). Legant and Bell (247) have considered that the "infarcts" are areas of necrosis due to vascular blocking with a reparative reaction in the living bone about the dead area This interpretation is based on the similarity to lesions in caisson disease and in patients following traumatic interruption of the circulation (247) The "infarcts" have also been attributed to capillary thrombosis in the bones (268) although this has not been demonstrated We have found no reports of histological study of these lesions by biopsy or at necropsy.

Disturbance in growth has been considered the cause of the observed abnormal dentition and delayed epiphyseal union observed (268, 375). Multiple bands of increased density near the epiphyseal ends of the long bones have been interpreted as indicative of periods of illness when growth ceased temporarily (268).

The roentgenological changes observed in sickle cell anemia are not specific and, with the possible exception of the translucent areas of bone that occur occasionally, may be seen in thalassemia or other hemolytic anemias When present, periosteal thickening and localized bone destruction may suggest syphilis, tuberculosis or leukemia. In older persons, the predominance of sclerotic bone reactions may lead to a mistaken diagnosis of osteoplastic carcinomatosis, Paget's disease or leukemia (114)

LABORATORY FINDINGS

Blood: Erythrocytes and Hemoglobin

Significant reduction in the erythrocytes and hemoglobin levels occurs in the vast majority of victims of the disease Erythrocyte counts as low as 950,000 per cubic millimeter (240) and 500,000 per cubic millimeter were also noted The average case of sickle cell anemia has approximately a 50 per cent reduction in both hemoglobin and erythrocytes Diggs (117) summarized the findings in 72 cases of sickle cell anemia and found the average erythrocyte count to be 2 6 million per cubic millimeter with the hemoglobin 45 per cent of normal The erythrocytes counts ranged from approximately 1 to 4 5 million while the hemoglobin varied from 10 to 90 per cent. Neel (301) reported an average erythrocyte count of 2 45 to 2.79 million per cubic millimeters for various age groups from 0 to 15 years

The hemoglobin average was 7.5 to 8.2 grams. Henderson (190) in a review of 31 cases found a hemoglobin range from 3 to 11 grams and red counts that varied from 1.2 to 4.0 million.

Table 2 shows certain hematologic data in our sickle cell trait and sickle cell anemia patients. In the patients with sickle cell anemia the erythrocytes varied from 1.2 to 4 8 million per cubic millimeter. There is a corresponding range of the hemoglobin from 4.1 grams to 13.0 grams

Anderson and Ware (15, 16) noted that the anemia is progressive at times while Josephs (217) considered it a rather striking feature of the disease that individual cases maintained a relatively fixed level of erythrocytes and hemoglobin Figure 2 demonstrates the red cell count and hemoglobin values obtained from frequent observations on a ten-year-old colored boy over a two year period A rather constant level was maintained except on two occasions at which time multiple transfusions were given. These values could be increased by such methods but following disappearance of the effects of transfusions there was a return to the pretransfusion level

The hematocrit in sickle cell anemia is decreased in proportion to the erythrocyte count and the hemoglobin. Averages of 37.6 per cent (117), 22.6 per cent, and 24 5 per cent have been reported (301). The values for mean corpuscular volume and mean corpuscular hemoglobin content are usually normal. In some patients with severe anemia a macrocytosis has been noted *(123, 470)*, and in others, a microcytosis *(162, 470)*. *In one* series of 44 cases studied by Diggs and Bibb (123) the average MCV found was 90 cubic microns In the same series, there was an average of 29 micromicrograms for the MCH and 32 per cent for the MCHC The mean cell diameter was 8 7 microns for cells of normal shape. Almost identical results were obtained in another study which showed an MCV of 85 9 to 96 7 cubic microns, MCH 29 1 to 33 1 micromicrograms, and MCHC 32 8 to 35 2 per cent (301). These reports indicate that the anemia is usually normochromic and normocytic

On a stained smear the erythrocytes occasionally assume the sickle form but more often only marked poikilocytosis and anisocytosis are present Most of the cells are of normal size and shape but macrocytes and microcytes may be observed along with bizarre cells which are usually elongated The long diameter of these cells may be such that the cell actually assumes a filamentous appearance Target cells are common Polychromatophilia of the diffuse type is common while the punctate *variety is less common* Cabot rings and Howell-Jolly bodies occur (100) and nucleated red cells are often present Sydenstricker (417) called attention to the fact that they may be present when sickle cells are few in number and difficult to demonstrate. In addition, he reported that they may increase preceding or accompanying a remission. These cells may number only 1 or 2 per 100 leuko-

cytes but may be much more numerous. In one of our patients they exceeded the number of leukocytes and on several occasions numbered over 80,000 per cubic mm. The number may vary markedly from day to day. In addition to true nuclei, a number of the erythrocytes contain nuclear fragments and occasionally sickled nucleated red blood cells are seen in the peripheral blood (418). Phagocytosis of the red cells by both adult granulocytes and large mononuclear cells has been reported (213, 415, 418) and may be evident on a fresh preparation of blood.

With proper staining increased numbers of reticulocytes can be demonstrated in the peripheral blood of practically all patients. The number varies with the individual and the stage of the disease, but when active manifestations of the disorder are present, it is not unusual to find counts of 20 per cent more. Diggs (117) found an average reticulocyte count o

TABLE 2

Hematologic Data: Normal, Sickle Cell Trait, and Sickle Cell Anemia

DIAG	HB	RBC	RETIC	S B	F U
	gm	mil	%	mg	mg /100 gm
Normal	15.0	5.0	0 3	0.6	30-200
S C Tr	13.0	4 9	0 2	0 4	26-106
SCA	13 0	4.8	5 3	2 1	260-900
SCA	8.4	2 8	9 0	3 8	1520
SCA	7 3	2 9	4 1	1 8	520
SCA	4 6	1.6	4.1	1 3	560
SCA	4.1	1 2	36.0	7 0	1120

15 per cent in 32 patients and was impressed by the fact that reticulated red cells in sickle cell patients did not sickle as readily as non reticulated ones It has been reported that a higher reticulocyte count may be found during remission than during active stage of the disease (132). Both the transfusion of normal blood and oxygen administration reduce the number of these cells in the circulation (245, 339, 340) Although reticulocytosis is the rule in the disease, a value as low as 0.2 per cent has been observed during an "aplastic crisis" (390).

Methods for Demonstration of the Sickling Phenomenon

Sickle cells were first observed in the blood by Herrick (195) in "fresh specimens, no matter in what way the blood was spread on the slide and they were seen also in specimens fixed by heat, alcohol, and ether, and stained with the Ehrlich triacid stain as well as with control stain" Emmel (137) first described the moist sealed cover slide preparation which has been employed by most students of the disease as the method of choice for

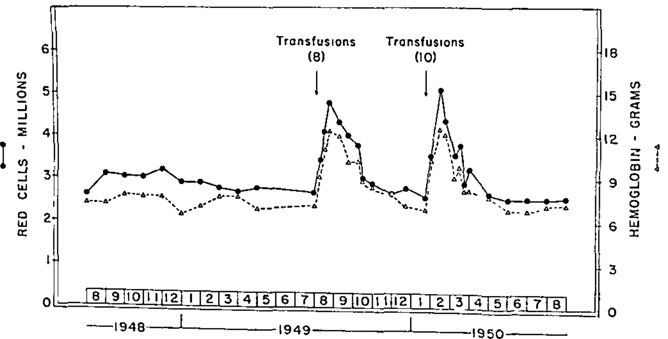


FIG. 2. F. G., SICKLE CELL ANEMIA

demonstration of the sickling phenomenon. Emmel's first description of his so called "culture preparation", made in 1917, appears adequate today "A ring of petrolatum was drawn on a carefully cleaned glass slide. A clean sterile cover glass was then brought in contact with a drop of fresh blood and the cover glass quickly placed on the slide in such a way to bring the drop of blood to the surface of the vaseline ring. The edges of the cover glass were also sealed with sterile petrolatum so as to form an air-tight chamber similar to that used for tissue culture purposes " Routinely this preparation is checked immediately and then at one-half hour, four hours, 24 and 48 hour intervals for the presence of sickle cells.

Other methods of demonstrating the sickling property have been described but none has enjoyed the same popularity as the simple sealed cover slip preparation of Emmel. It is known that the hemoglobin in sickle cells must be in the reduced state in order for the sickling phenomenon to occur (39, 61, 171). All of the tests described have been based on this fact. The tests are listed in table 3

Of the tests available, the cover slip preparation of Emmel is the simplest and least tedious method, but, unfortunately, not entirely reliable The main weaknesses of this method are its high incidence of false negative reactions and its variability. The results frequently vary from day to day (14, 101). Even two separate preparations on opposite ends of the same slide may differ, revealing sickle cells in one but not in the other A difference in the size of the cover slip may influence the result (158). An imperfect seal which allows oxygen to enter the preparation will delay sickling or even cause sickle-shaped erythrocytes to assume the discoid form Some of the other methods are cumbersome and require special equipment. The most practical methods are those which employ sodium hydrosulfite, cevalin, sodium bisulfite BAL, cysteine, and hydrogen sulfide. Even these methods have the disadvantage of requiring special reagents which, with the exception of hydrogen sulfide, are unstable and must be prepared fresh each day for the best results. These tests are of special value in large surveys where groups of individuals are to be checked for the sickling trait

In comparing the stasis, non stasis (Emmel) and formalin fixation methods, Diggs and Petitt (125) noted positive results in 36 instances with the stasis method in 37 known carriers of the sickle cell trait The formalin fixation method was positive in 35 while the non stasis method was productive of only 31 instances of sickling In survey work, Neel (301) utilized several methods, the venous stasis method (368), the Janus Green method (181), the ascorbic acid (221) and formalin fixation (316). The formalin fixation method appears best for the permanent preparations

Sedimentation Rate

The sedimentation rate in sickle cell disease is not as elevated as one would expect in a patient with so much anemia. Even with very low erythrocyte counts and hemoglobin values, the erythrocyte sedimentation rate may be normal The sedimentation rate is not retarded in all patients (158, 287). Inhalation of pure oxygen accelerates the sedimentation while rebreathing into a paper bag retards it (467). Prolonged stasis of venous blood by a tourniquet around the extremity from which the blood is drawn will also cause retardation (467) In the same sample, exposure of blood to carbon dioxide gas will retard the sedimentation rate to less than 1 mm. per hour while oxygenation of the blood will cause it to increase from 23 to 70 mm in the same length of time (61). These observations indicate that the sedimentation rate is related to the form of the erythrocytes and the results of several tests cannot be compared unless the degree of oxygenation was comparable. The behavior of the sedimentation rate in this disease is the basis for an indirect method of demonstrating the sickling property When postive, this test, which has been designated as the "diagnostic parameter" is highly indicative of the presence of the sickling phenomenon (322, 463).

TABLE 3

Methods Used to Demonstrate the Sickling Phenomenon

AUTHOR	YEAR	METHOD
Emmel (137)	1917	Moist sealed cover slip preparation
Scriver & Waugh (368)	1930	Venous stasis
Beck & Hertz (32)	1935	Formalin fixation
Hahn & Gillespie (172)	1927	CO_2 gas chamber
Hanno & Margolies (180)	1950	CO_2 flask
Hansen-Pruss (181)	1936	Brilliant cresyl blue
Hansen-Pruss (181)	1936	Janus green
Hansen-Pruss (181)	1936	Methylene blue
Hansen-Pruss (181)	1936	Sodium cyanide
da Silva (382)	1948	Sodium hydrosulfite
Itano & Pauling (210)	1949	Sodium bisulfite
Daland & Castle (110)	1948	Ascorbic acid
Thomas & Stetson (422)	1949	Hydrogen sulfide
Thomas & Stetson (422)	1949	BAL
Thomas & Stetson (422)	1949	Cysteine
Neuda & Rosen (304)	1945	Feces-broth culture
Singer & Robin (391)	1948	Exposure of the erythrocytes to bacteria

Erythrocyte Resistance to Trauma and Saline

Abnormal resistance to osmotic and mechanical factors has been shown to be a property of the sickle cell. Simple mechanical shaking is ineffective in causing destruction of sickle cell anemia erythrocytes unless the blood is first treated with carbon dioxide or removed from oxygen by exposure to a nitrogen atmosphere (69). Under such circumstances, the erythrocytes assume the distorted forms and show marked susceptibility to mechanical trauma. Increased resistance to hypotonic saline is the usual finding in the disease (17, 117, 123, 158, 278, 287, 398) In 25 cases, Diggs (117) reported an average initial hemolysis at 0 35 per cent with complete hemolysis at 0.22 per cent as compared with the normal values of 0 45 to 0.39 per cent for initial hemolysis and 0 33 to 0 30 per cent for complete hemolysis (470). Levels as low as 0 09 per cent (287) and 0.12 per cent (158) have been reported for complete hemolysis and the survival of some cells in distilled water has been reported (123). Rarely fragility has been found increased (398). Sickle cell trait blood has been reported to show a similar increase in resistance (123) while others report that sicklemia cells behave normally (101).

Leukocytes

The leukocyte count in the majority of patients usually is elevated even after a correction is made for the circulating nucleated red cells. The average leukocyte count was 18,000 per cubic mm. in one series (117) Leukopenia was not found in this series In other series, averages of 12,820 to 16,170 per cubic mm for several age groups have been found (301), and ranges from 4,400 to 48,000 per cubic mm. observed (190). Total counts as high as 54,000 to 69,000 (355) have been reported but in both instances there were complicating diseases that may have affected the leukocytes A decrease in the white count is observed during partial remissions in the course of the disease At that time the leukocytes may reach normal levels, although more often some elevation persists. With even mild exacerbations a prompt rise usually occurs.

In addition to an elevated leukocyte count, the Schilling differential count reveals increased number of bands, juveniles, and occasionally myelocytes along with an increase in the mature segmented forms A leukemoid picture with marked leukocytosis and predominance of myelocytes and metamyelocytes has been reported (355). A slight increase in eosinophiles has been reported by some authors (278, 415) but not by all (117). Concentrates of the leukocytes in the peripheral blood do not reveal any unusual finding.

Platelets, Bleeding and Clotting Times

The platelets counts are at the upper limit of normal or higher. An average count of 369,000 per cubic mm. was found by Diggs (117). Some of our patients have had counts of over a million. The bleeding and clotting times are generally normal and no abnormalities are noted in the structure on fragility of the clot.

Blood Chemistry

The plasma and serum proteins have been studied by electrophoretic methods In one study of 15 patients with severe sickle cell anemia (31), there was hypoalbuminemia in 13, elevated beta globulin in 3, and reversal of the A/G ratio in 12. These non-specific changes were thought to be due to the general disease process, especially in the liver, caused by the sickled cell. One Negro with sickle cell trait had a normal protein pattern, and three patients under eight years of age had only minor abnormalities in the plasma

Plasma iron levels of from 0.041 to 0.215 mg. per 100 ml have been reported (339). In eight of our patients with sickle cell anemia, the plasma iron levels varied from 0 035 mg per 100 ml. to 0 260 mg. per 100 ml. The values varied greatly in different patients, and from time to time in the same patient. Subnormal values occurred only during infections, or in women who menstruated

The free erythrocyte protoporphyrin was determined repeatedly in the same eight patients All values were normal or increased and ranged from 33 to 360 mg In general, the patients with the most severe anemia and most marked reticulocytosis had the greater elevations, but periodic determinations of the free erythrocyte protoporphyrin in the same patients revealed variations independent of the variations in the reticulocyte count, degree of hemoglobin destruction, changes in the erythrocyte count, or the presence of infection

The serum bilirubin and icterus index are usually elevated Whereas the icterus index usually is not over 30 units, values of 150 and 280 units have been recorded (313) in two patients with bilirubinuria in whom hepatitis and cholecystitis were suspected but not proved In another series of 51 patients (190), 20 had an elevated direct reacting bilirubin, 20 had an elevated delayed-reacting bilirubin, and 11 had values within normal limits. In a small series of eight patients that we have followed regularly, the plasma bilirubin values varied from 0 7 to 7 5 mg The highest figure, which included 3.7 mg. of the 1 minute direct-reacting bilirubin, occurred in a patient who was severely ill in crisis with obvious liver damage As in other hemolytic anemias, occasionally the serum bilirubin was normal

(0 7 mg.) when the fecal urobilinogen was definitely elevated (480 mg./100 gm). In crisis the serum billirubin usually rises but not always.

In a series of 13 cases, the blood cholesterol ranged from 208 to 877 mg per 100 cc. while the majority had values between 260 and 330 mg. per 100 cc. (158). These same authors noted blood calcium values from 6 0 to 11 8 mg. (average 8 33 mg.) in 14 patients. Inorganic phosphorus ranged from 2 04 to 6.45 mg. in 18 individuals.

Bone Marrow

Preparations of the bone marrow made during life reveal it to be quite active and similar to that found in other types of hemolytic anemia. The appearance of the bone marrow has been described (346). There is an increase in the early cells of the granulocytic series and a marked increase in the normoblastic series. No abnormal cells other than sickled erythrocytes are found in the bone marrow preparation. In addition to sickled mature erythrocytes, it is not unusual to observe sickled nucleated red cells Some erythrocytes become elongated and filamentous and present a bizarre picture when interspersed among the normal erythrocytic and granulocytic elements. At times, these filamentous forms reach a length that may equal the diameter of the oil immersion field but the majority are from a fourth to one half this length. On the basis of an autopsy in one patient, it has been stated that bone marrow exhaustion is probably reached beginning in the third decade in those who survive to this age (158). This finding must be the exception rather than the rule, for we have seen patients beyond this age who had a hyperplastic marrow and no evidence of increasing anemia Acute transient marrow aplasia has also been reported in crisis that developed during an attack of pneumonia (390)

Urine

The occurrence of albumin, cells, and casts in the urine in this disease has been noted frequently The urine is often deep brown in color as a result of the increase in urobilinogen (157, 201). Ehrlich's benzadehyde reagent may reveal increased amounts of urobilinogen but the increase is not always found The specific gravity is usually normal, but has been observed to become fixed at a low level (157, 201)

Stool

Routine examination of the stool reveals no significant abnormalities. Quantitative determinations of the fecal urobilinogen reveal it to vary from normal to a marked elevation Determination of 40 single specimens obtained from 10 patients with sickle cell anemia have revealed values that

ranged from 160 to 2,640 mg./100 grams (246). In only 2 instances were values of less than 250 mg /100 grams obtained. Differences of more than 700 mg./100 grams in the random specimens occurred without significant changes in the erythrocyte count or plasma bilirubin Average total daily fecal urobilinogen excretions of between 200 and 750 mg have been reported (339, 340). The fecal urobilinogen may decrease spontaneously at times during the course of the disease, and a decrease after transfusion of whole blood (245), and after plasma (224) has been reported. Administration of oxygen in high concentration produced no consistent effect (339).

Serology

Unless there is coincidental infection with syphilis, the Wassermann and Kahn reactions are usually negative but development of a positive Wassermann and Kahn during crisis has been described (480) Whether this case represented syphilis or a false positive reaction that developed during exacerbation of symptoms is not clear.

Cerebrospinal Fluid

There are no specific changes in the cerebrospinal fluid referable to sickle cell anemia. When cerebral hemorrhage occurs, the findings then do not differ from those expected from hemorrhage due to any other cause Red cells in varying numbers, even grossly bloody spinal fluid, may be present, associated with an elevation of protein and spinal fluid pressure Sickled red blood cells may be noted in the fluid obtained at spinal puncture (203, 223)

Pathology

Comprehensive articles devoted to the detailed pathological aspects of the disease have been written by Diggs and Ching (124) and Mathews (279) Special articles have reported the changes that are found in the spleen (121, 341, 396), in the brain (55, 203, 457), in the heart (233), and in the skeleton (126). The histological appearance of the ulcers has been described (109, 124, 201, 440) The pathogenesis of the lesions has been discussed in several papers (124, 229, 426) Autopsy reports, which vary in point of interest and in degree of completeness, are fairly numerous (15, 30, 38, 92, 104, 164, 179, 85, 188, 189, 157, 201, 273, 326, 365, 447, 474, 476, 477, 229, 415). Other articles of a summary nature have included a discussion of the pathology (109, 159, 406, 470).

The most striking feature of the pathology of the disease is the presence of the sickled erythrocytes, which are demonstrated better in tissues fixed with formaldehyde than in those fixed with Zenker's solution (124, 159). The degree of distortion varies in different patients, in different organs in

the same patient, and in different blocks of the same tissue (124). The next most conspicuous feature is capillary engorgement common in all tissues (124) Associated with the occurrence of deformed erythrocytes in the congested capillaries, lesions of two general types occur. Areas of infarction are often found in the various organs, and these have been attributed to thrombosis (124, 406, 477) and to ischemic necrosis (229). The areas of fibrosis are thought to be a late result of the infarction. The other group of lesions includes those associated with increased blood destruction. In this category are the pigment deposits that are found in the liver, bone marrow, lymph nodes, spleen and kidney and the phagocytosis of erythrocytes by the reticuloendothelial system The marrow hyperplasia appears to be a response to the excessive hemolysis.

The gross examination at autopsy may reveal nothing suggestive of sickle cell anemia At other times, the tall slender build, the high skull, the tapering digits, poorly developed genitalia, pale mucous membranes and leg ulcers may suggest the diagnosis Generalized adenopathy of moderate degree is often present (124) The reports of the lesions that have been observed in the individual organs have been included in the discussions of the "Mechanism of Disease" and the various "Clinical Manifestations".

ASSOCIATION WITH PREGNANCY

Many patients with sickle cell anemia survive to the child bearing age. We have found reports of the occurrence of pregnancy in 84 patients with sickle cell anemia (table 4) The incidence of sickle cell anemia in Negro deliveries has been reported as 1:5,000 (277), 1:1296 (13), 0:10,000 (31). In all probability the association of pregnancy and sickle cell anemia occurs more often than is reflected in the case reports, since not all of the recognized instances have been reported, and undoubtedly many have passed unrecognized unless tests for sickling have been done routinely on Negro obstetrical patients The published reports suggest that the prognosis for the mother with sickle anemia and for the fetus is poor as compared to the normal The presence of the sickle cell trait does not interfere with pregnancy in any way (414).

The association of pregnancy and sickle cell anemia has been discussed in some detail in several papers (13, 31, 80, 151, 235) and has been the subject of case reports in others (112, 145, 194, 242, 253, 271, 277, 291, 307, 343, 374, 402, 481)

These reports, which have been summarized in table 4, present a very unfavorable picture for both the mother and the child In the total of 150 pregnancies, there were 17 abortions, an incidence of 13 per cent. In 80 patients reported in enough detail to be evaluated, there were 14 still births

(17 per cent) and 9 post partum deaths (12 per cent), for a total fetal mortality, excluding abortions, of 28 per cent There were 16 maternal deaths in the 81 patients in whom the outcome was known, mortality rate

TABLE 4
Sickle Cell Anemia and Pregnancy

CASE	REFERENCE	YEAR	AGE OF PT	NO PREV PREGNANCIES	MATERNAL COMPLICATIONS		FETAL DEATHS
					Death	Other	
1, 2	(415)	1924	—	—	—	—	—
3	(477)	1931	25	1	1	—	1
4	(242)	1934	21	0	1	—	1
5	(342)	1934	16	0	0	0	0
6	(374)	1936	26	—	0	0	0
7	(228)	1936	28	6	1	—	—
8	(253)	1937	25	3	0	0	1
9	(401)	1938	23	2	0	0	0
10–11	(314)	1939	19–20	0	1	—	1
12–17	(235)	1940	18–26	6	1	3	3
18	(441)	1943	26	1	—	—	0
19	(404)	1945	35	0	0	0	0
20	(307)	1946	32	Yes	1	—	1
21	(481)	1946	20	0	0	0	0
22–23	(277)	1947	16–18	0	0	0	0
24–29	(80)	1947	18–33	9	1	3	2
30–32	(200)	1947	21–23	1	0	3	1
33	(166)	1947	17	0	0	1	1
34	(299)	1948	21	0	0	0	0
35	(145)	1948	23	3	0	1	1
36	(128)	1948	32	2	0	1	0
37	(343)	1949	17	0	1	0	1
38	(112)	1949	21	1	0	0	0
39–44	(151)	1949	16–25	8	4	0	2
45–55	(13)	1949	17–29	6	0	4	4
56	(254)	1950	32	0	0	0	0
57	(271)	1950	32	4	0	0	0
58	(70)	1950	21	0	0	1	0
59–60	(402)	1950	24	0	0	0	0
61–84	(31)	1950	16–32	13	4	"46"	"40"

of 20 per cent Even if the mortality rate is calculated on the basis of 150 pregnancies, it remains a formidable 10 6 per cent

As striking as the mortality figures are, they do not give the complete picture of the difficulties that arise during pregnancy, delivery and the post partum period Complications during labor are not common, but

pelvic abnormalities, which have been demonstrated by x-ray pelvimetry, may be responsible for difficult labor (13). Manifestations of toxemia of pregnancy such as headache, convulsion, hypertension and edema are common (13, 31, 145, 200, 307). Admission to the hospital may be necessary because of crisis, or because of severe anemia. Even the use of transfusion may be associated with increased hazard because of an apparently high incidence of transfusion reactions (13), and the frequency of coexisting heart failure (151). The most frequent cause of maternal death is said to be heart failure (150) but infection (31, 80, 151, 229), pulmonary embolism (314), uremia (80), shock (343) and cerebrovascular accidents (343) may also be responsible.

It has been stated that the properly handled obstetrical patient with sickle cell anemia is in no more serious difficulty than patients with other chronic diseases (402). In one series of 11 patients, there were no maternal deaths (13). Thus, while the outlook for these patients is not invariably gloomy, the mortality rate of about 20 per cent for the mother and about 40 per cent for the fetus in the reported cases means that careful observation and prompt treatment are particularly necessary in this group of patients. It is possible that more information about this problem, information that can be gained only through a routine search for sickle cell anemia in all Negro obstetrical patients, may modify the present picture. The reported results may be influenced by the tendency of patients with mild sickle cell anemia and those without complications to pass undetected and unreported, while those who develop unexpected complications attract attention.

Some observers have thought that a lower degree of fertility exists in women with sickle cell anemia as compared to the normal (112, 401). Two possible explanations have been offered, an effect of the anemia per se or the result of some other aspect of the disease. Anemia of even mild degree has been considered to produce considerable depression of spermatogenesis and, presumably oogenesis (282) If anemia exerts such an effect, it should be apparent in sickle cell anemia, where the anemia is so constant over a period of years. Other observers do not agree that there is a reduction of fertility in sickle cell anemia (163, 169). The lowered incidence of sickle cell anemia that is reported in pregnant women 1:1296 (13), 1:1550 (31), 1:5000 (277) is very difficult to evaluate It has been thought to be low, because of the incidence of sicklemia of about 9 per cent (275), and an estimated incidence of one sickle cell anemia to forty individuals with the sickle cell trait. This would give an incidence of about 2.25 per thousand Negroes (122, 301) The exact influence of age on these frequencies is unknown but it seems probable that the mortality in sickle cell anemia is appreciable before maturity is reached Thus, it is possible that what appears

to be a low incidence of sickle cell anemia in pregnant women, may be the same as the incidence of the disease in all individuals in that age group.

THE QUESTION OF CLINICAL SYMPTOMS WITH THE SICKLE CELL TRAIT

In the early history of sickle cell disease, no distinction was made between sickle cell anemia and sickle cell trait without anemia Later it was recognized that many individuals with the sickling trait had no anemia and appeared normal in all respects except for the sickling characteristic. This led to the concept that sickle cell anemia was a severe disorder while sickle cell trait was an inherited defect of the erythrocytes that was interesting but harmless. This belief, which was accepted generally for many years, has been challenged recently by an increasing number of reports of patients with the sickling trait who developed serious manifestations of the disease even though anemia was absent

It has been recognized that varying degrees of anemia occur in sickle cell anemia, that mild degrees of anemia may be associated with symptoms, and that mild degrees of anemia may not be recognized for various reasons. It is not so generally recognized or accepted that the manifestations of the disease may occur in patients whose blood is normal except for sicklemia. If the differences between the cells of sickle cell trait and sickle cell anemia is one of degree only, and if serious manifestations of the disease are the result of the assumption of the abnormal sickle form in vivo, it would not be surprising for patients with the sickling trait to develop manifestations of the disease in severe anoxia, such as might arise in shock, during anesthesia or certain disease states. That such developments do occur is the interpretation of authors who have reported what they considered to be examples of this type However, there is considerable difference in the implication if the patients in question have "severe symptoms without persistent anemia" as reported by Green and Conley (162), or "sickle cell disease without anemia" as reported by Bauer (28, 30) and Pratt-Thomas and Switzer (334). This matter is of such importance clinically, that a careful and critical appraisal of the case reports was undertaken

The report of Levy in 1929 (250) concerned 12 patients with sicklemia found on 213 hospital admissions None had marked anemia but 8 had hemoglobin levels that ranged from 60 to 80 per cent Three others had hemoglobin values of 85, 95 and 96 per cent, but there is nothing to indicate that these patients with sicklemia had symptoms at any time

The report of Bauer and Fisher (30) did not contain many details which are necessary in order to establish the cases as instances of sickle cell trait without anemia. In case 1, the hemoglobin was "90 per cent", during an episode of abdominal pain and vomiting seven months before death No other blood counts were reported At autopsy the spleen weighed 550

grams, and it was considered that no anemia existed because at autopsy "no anemia was found". The dark purplish color of the liver was considered not to be in accord with the anemia Cases 2 and 3 had no determinations of the hemoglobin or erythrocytes, but anemia was considered improbable on autopsy because the cut sections of the livers were "mahogany mottled brown" and "brownish-black", although one spleen weighed 620 grams and the other 25 grams The abnormalities of the spleen noted in these cases and the description given of the liver are those often found in patients with sickle cell anemia. Cases 4, 5 and 6 in this report had anemia Case 7, which was reported in detail by Conley, Carpenter and Elmer (79), had only a leukocyte and differential count prior to splenectomy. Studies done at uncertain time after splenectomy and transfusions revealed the icterus index to be 44, erythrocyte count 4.7, hemoglobin 12 grams, leukocyte 24,000, and the nucleated red cells numbered 5 per 100 leukocytes. The conclusion which the authors reached in these cases, that severe sickle cell disease existed without anemia, does not rest on very strong evidence.

Pratt-Thomas and Switzer (334) reported an interesting group of 10 patients who died suddenly, some for unknown reasons, after operation The main pathological finding was profound sickling It was concluded that sicklemia is at times responsible for untoward developments in the presence of the lowered oxygen tension In their report, detailed blood studies were not given but case 1 had a hemoglobin of 12 grams, case 2, 11 grams; case 3, a hemoglobin of 78 per cent and case 6, 11 grams. In cases 5, 7, 8, 9 and 10 no blood studies were reported In case 4, a 26-year-old man, who was acutely ill, mentally confused, and had a fever of 102° for three days, the hemoglobin was 16 grams; profound shock and death occurred after incision and drainage of a submental abscess under local anesthesia. Autopsy revealed sickling of erythrocytes which formed agglutinated masses in many blood vessels, and early lobar pneumonia The conclusion that death was due to sicklemia without anemia does not appear justified by the data.

Thompson, Wagner and MacLeod (424) reported a 20-year-old Negro man who died after an acute febrile illness with evidence of disease of the central nervous system The hemoglobin was 15 grams, the erythrocytes, 4 9 million Autopsy revealed laminated thrombi in the cortical veins, which resulted in intense congestion of the cortical veins and capillaries, diffuse cortical hemorrhages and bleeding into the subarachnoid space The role of the sickled cells in this case is hard to assess, but the demonstration of sickling in areas of hemorrhage at necropsy does not necessarily mean that thrombosis and hemorrhage were the results of the sickling

Green and Conley (162) reported three interesting patients who had clinical manifestations of sickle cell disease and anemia although the latter was mild and microcytic in two and inconstant in the other. They were

to be a low incidence of sickle cell anemia in pregnant women, may be the same as the incidence of the disease in all individuals in that age group.

THE QUESTION OF CLINICAL SYMPTOMS WITH THE SICKLE CELL TRAIT

In the early history of sickle cell disease, no distinction was made between sickle cell anemia and sickle cell trait without anemia. Later it was recognized that many individuals with the sickling trait had no anemia and appeared normal in all respects except for the sickling characteristic. This led to the concept that sickle cell anemia was a severe disorder while *sickle cell trait was an inherited defect of the erythrocytes that was interesting* but harmless. This belief, which was accepted generally for many years, has been challenged recently by an increasing number of reports of patients with the sickling trait who developed serious manifestations of the disease even though anemia was absent.

It has been recognized that varying degrees of anemia occur in sickle cell anemia, that mild degrees of anemia may be associated with symptoms, and that mild degrees of anemia may not be recognized for various reasons It is not so generally recognized or accepted that the manifestations of the disease may occur in patients whose blood is normal except for sicklemia. If the differences between the cells of sickle cell trait and sickle cell anemia is one of degree only, and if serious manifestations of the disease are the result of the assumption of the abnormal sickle form in vivo, it would not be surprising for patients with the sickling trait to develop manifestations of the disease in severe anoxia, such as might arise in shock, during anesthesia or certain disease states. That such developments do occur is the interpretation of authors who have reported what they considered to be *examples of this type However, there is considerable difference* in the implication if the patients in question have "severe symptoms without persistent anemia" as reported by Green and Conley (162), or "sickle cell disease without anemia" as reported by Bauer (28, 30) and Pratt-Thomas and Switzer (334) This matter is of such importance clinically, that a careful and critical appraisal of the case reports was undertaken.

The report of Levy in *1929 (250)* concerned *12* patients with *sicklemia found* on 213 hospital admissions None had marked anemia but 8 had hemoglobin levels that ranged from 60 to 80 per cent Three others had hemoglobin values of 85, 95 and 96 per cent, but there is nothing to indicate that these patients with sicklemia had symptoms at any time.

The report of Bauer and Fisher (30) did not contain many details which are necessary in order to establish the cases as instances of sickle cell trait without anemia In case 1, the hemoglobin was "90 per cent", during an episode of abdominal pain and vomiting seven months before death. No other blood counts were reported. At autopsy the spleen weighed 550

cell anemia but it is rarely found in Negroes (470). Distinction should not be difficult since the spleen is enlarged more consistently, microspherocytes occur, and the erythrocytes show a decreased resistance to saline solutions rather than the increased resistance of sickle cell anemia. Of equal importance is the differential diagnosis of thalassemia major (Cooley's anemia, Mediterranean disease). Splenomegaly and the mongoloid facies are common, the anemia is typically hypochromic microcytic and the sickling test is negative Other hemolytic states such as those associated with leukemia, Hodgkin's disease, carcinomatosis, liver disease, infections, chemical agents, poisons, and abnormal antibodies, offer problems, but the clinical course associated with these disorders usually suffices to distinguish them from sickle cell anemia. The Coombs test might be helpful The sickle cell test is usually negative but it is conceivable that these states may occur in individuals with the sickle cell trait. Under these circumstances, diagnosis might prove difficult Pernicious anemia should be easily differentiated from sickle cell anemia by the finding of a macrocytosis, achlorhydria and a megaloblastic bone marrow. The chronic hypochromic anemias from chronic blood loss often exhibit variations in the shape of the cells, but confusion with the cells of sickle cell anemia is unlikely. Usually reticulocytes comprise less than 5 per cent of the non-nucleated erythrocytes (273), the icterus index is normal, bone changes are absent, and splenomegaly is rare.

In certain patients, the cardiovascular manifestations of sickle cell anemia may be confused with cardiovascular disease of other etiology, especially rheumatic fever. Certain features may help to distinguish between the two conditions The extremity pain in sickle cell anemia usually is not limited to the joints but involves other parts of the extremities. Actual signs of joint involvement such as heat, swelling, and tenderness are less frequently observed in sickle cell anemia Response to salicylate therapy is seldom dramatic in sickle cell anemia as it often is in acute rheumatic fever The simultaneous occurrence of pain in other regions of the body, especially in the abdomen and chest, occurs more often in sickle cell anemia Left auricular enlargement is rare in sickle cell anemia and murmurs, while consistently present, are usually systolic rather than diastolic in time. A changing P-R interval is more suggestive of rheumatic fever. The sedimentation rate, which is characteristically elevated in rheumatic fever, may be normal or only moderately elevated in sickle cell anemia Under certain circumstances, other forms of heart disease such as bacterial endocarditis, coronary sclerosis and congenital manifestations may be suspected but the correct diagnosis is usually established without difficulty after proper study

Signs of acute intra-abdominal disease occur frequently in sickle cell anemia Since patients with sickle cell anemia may have either acute surgi-

also unusual in that all showed microcytosis, which further suggested a similarity between sickle cell anemia and thalassemia.

A review of the reported cases and the discussions on the subject leads to the conclusion that, while the occurrence of symptoms from the sickle trait alone might occur under proper circumstances, the existence of such cases has not yet been demonstrated beyond reasonable doubt. Symptoms undoubtedly occur in patients with mild anemia and other abnormalities of the blood such as mild reticulocytosis, microcytosis and increased fecal urobilinogen excretion that may not be apparent on routine blood examinations. Unfortunately, if the anemia is not severe, the possibility of sickle cell disease often is not considered until necropsy reveals the characteristic sickling. In any patient with symptoms suggestive of sickle cell disease, tests for sickling are indicated. If sicklemia is present, careful and detailed study of the blood and hemoglobin metabolism should be made. If these studies are entirely normal, and no other explanations for the symptom is found, one might then suspect that clinical symptoms have occurred as a result of the sickle cell trait None of the reported cases has met these requirements In view of the frequent occurrence of the sickle cell trait in Negroes, if this abnormality were responsible for clinical manifestations, one would expect such developments to be much more frequent than the few reports to date would indicate. Certainly many have gone through pregnancy, anesthesia, infections, and shock, without any unusual complications The report of Green and Conley (162) is timely since it emphasizes the importance of recognizing patients with mild degree of anemia.

DIFFERENTIAL DIAGNOSIS

If tests for sickling are done on all Negro patients, particularly those who present the picture of a generalized disease with anemia, the diagnosis of sickle cell anemia rarely presents any difficulty When other forms of anemia develop in an individual with the sickle cell trait, difficulty may arise. We have seen a sicklemic person with pernicious anemia and others with anemia due to chronic blood loss, who were followed for considerable periods in the clinic under the incorrect diagnosis of sickle cell anemia A positive sickling test means that the individual has sicklemia, but the coexistence of a positive test and anemia does not warrant a diagnosis of sickle cell anemia The diagnosis can be established by demonstrating other characteristics of the state such as reticulocytosis, leukocytosis, elevated serum bilirubin and increased fecal urobilinogen, if other types of hemolytic anemia are excluded. Should the tests for sickling be omitted, the disorder may be confused with a number of disease entities and pass unrecognized It has been called rightly "a great masquerader" (468)

Familial spherocytosis exhibits many features in common with sickle

correcting the anemia. Opinions have varied regarding the usefulness of transfusions, the indications for their use, and the objective to be attained. If the erythrocyte count is brought to normal by transfusions the anemia is abolished temporarily, and, at least in some patients, the number of cells that are subject to sickling is reduced markedly, to 10 per cent or less, within a few weeks (245). This suggests the need for evaluation of this form of treatment in the preparation of the patient for elective surgery or delivery, since the incidence of vascular accidents might be reduced. Unfortunately, the effects of transfusions disappear in a few months, and the danger of transfusion hemosiderosis would seem to preclude the use of multiple transfusions in an effort to maintain normal blood values as a form of replacement therapy for a prolonged period.

Splenectomy was suggested as a means of therapy first in 1924 by Sydenstricker (417). In 1927, Hahn and Gillespie reported the first instance of splenectomy (172). Since that time, a number of splenectomies have been performed in an attempt to alleviate the anemia. The results have been reviewed in detail recently (380). Although this operative procedure has no place in the treatment of the usual patient with sickle cell anemia, it merits consideration in those with marked splenomegaly, since significant improvement has followed splenectomy in some patients of this type.

Probably the most encouraging report on therapy is that of Kass, Ingbar, Harris and Ley (225) They reported on the use of ACTH in two patients. In one, typical crisis occurred following the administration of ACTH, but the hematologic data showed no change In the other patient, after an initial episode of crisis, continued use of the hormone was accompanied by a rise of the erythrocytes to normal, a diminution in reticulocytes and sickled forms, and a return of the osmotic and mechanical fragility to normal Relapse occurred when the hormone was discontinued. The authors thought that the effect of ACTH was the result of its ability to alter sulfhydryl activity within the erythrocytes Prior to the report of these authors, we treated one patient with ACTH, and found that crisis occurred on three occasions when the hormone was given No change was observed in the erythrocytes, plasma bilirubin or plasma iron Further study is necessary before this form of treatment can be evaluated. Only normal adrenal glands have been reported at autopsy (157, 476)

SUMMARY

Sickle cell anemia is an inherited disorder apparently confined to Negroes or persons with some Negro ancestry This inherited defect is responsible for the production in the erythrocyte of an abnormal type of hemoglobin. When this hemoglobin is in the reduced state, the erythrocyte assumes a characteristic "sickle" shape When the erythrocytes are in this form,

cal conditions that demand operative intervention, or a similar picture that results from the disease itself, correct diagnosis may be extremely difficult. This problem has been discussed ably in several papers (69, 316, 462). The possibility of sickle cell anemia should be considered in all Negroes who present the picture of an acute abdomen

Ulcers of the extremities must be differentiated from tuberculous ulcerations (erythema induratum and scrofuloderma), syphilis (gummatous syphiloderma or gumma), traumatic ulcerations (407), circulatory stasis, congenital hemolytic jaundice and coccidioidal granuloma (348). Blood and spinal fluid serological tests may exclude syphilis from consideration Where both syphilis and sickle cell anemia exist together, the diagnosis may be made only after a therapeutic test with anti-syphilitic drugs. Histopathologic examination, cultures and animal inoculation will usually aid in the problem of differentiating ulcers of other etiology.

When a cerebrovascular accident is the presenting symptom in a patient with sickle cell anemia, arteriosclerosis, syphilis, congenital vascular anomalies, infections and neoplasm of the central nervous system may be diagnosed. Other diseases with which sickle cell anemia has been confused include atrophic arthritis (93), osteomyelitis (120), poliomyelitis (156), and infectious hepatitis (12), bilharziasis, blackwater fever, yaws and yellow fever (140)

THERAPY

At the present time, the treatment of sickle cell anemia leaves much to be desired since it is limited to symptomatic and supportive measures Many manifestations of the disease such as congestive heart failure, cutaneous ulceration, priapism, cerebrovascular accidents, pain, and infections are treated in the same way as they are when they occur in other disorders

Efforts to reduce in vivo sickling and relieve vasocongestion have not been very successful Oxygen (28, 248, 432) and sodium bicarbonate (248) have been employed. Reinhard, Moore, Dubach and Wade (339, 340) have demonstrated that when high concentrations of oxygen are used, marrow activity decreases while no significant decrease in hemolysis occurs. Nitroglycerine, atropine, adrenaline (432) desiccated thyroid, warm enemata, douches, intravenous infusions, massage and exercise (28) are measures that have been suggested on theoretical grounds, but no effects on vasocongestion have been demonstrated

The preparations that have been used in an attempt to increase blood [illegible] such as liver diet (86, 250), liver extract (118), iron (86, 118), [illegible] pheral blood. Blood transfusions appear to be the most effective means of

8. ALMKLOV, J R., HANSEN, A. E. AND SCHNEIDER, M.: Long bone involvement in sickle cell anemia. Ped , 5: 204, 1950
9 ALONZO PATIÑO, M , LEON, L AND LOTT, L M : Anemia a hematies falciformes (sickle-cell); observacion de un caso en adulto joven Arch de. med. Int., 4: 220, 1938
10 ALTMAN, A.: Sickle cell anemia in South African born European. Clin Proc., 4: 1, 1945
11 ALTMAN, A.: The sickle cell trait in the South African Bantu. S. African Med. J., 19: 457, 1945
12 ALTMAN, A : The survival of transfused erythrocytes in sickle cell anemia Tr. Roy. Soc. Trop. Med. & Hyg , 40: 901, 1947.
13 ANDERSON, G. W. AND BUSBY, T . Sickle cell anemia and pregnancy Am J Obst. & Gyn , 58: 75, 1949
14. ANDERSON, H B : Sickle cell anemia report of an active case. Am J Med Sci , 171: 641, 1926.
15 ANDERSON, W. W. AND WARE, R. L.: Sickle cell anemia Am J Dis. Child , 44: 1055, 1932
16 ANDERSON, W W. AND WARE, R L Sickle cell anemia. J. A M A , 99: 902, 1932
17. ANDERSON, W W. AND WARE, R L Sickle cell anemia. Tr , Sect on Pediat , A M. A , 97: 99, 1932.
18 ARCHIBALD, R G A case of sickle cell anemia in the Sudan Tr. Roy. Soc. Trop Med. & Hyg , 19: 389, 1925–26
19 ARENA, J. M : Cerebral vascular lesions accompanying sickle cell anemia J Pediat , 14: 745, 1939
20 ARENA, J M : Vascular accident and hemiplegia in a patient with sickle cell anemia Am J Dis Child , 49: 722, 1935
21. ASHBY, W. J : Determination of the length of life of transfused blood corpuscles in man J. Exp Med , 29: 267, 1919
22 AUER, J The structure and function of filaments produced by living red corpuscles Am J Med Sci , 186: 776, 1933
23 BAIRD, J A Sickle cell anemia report of a case with multiple infarctions and necropsy M Bull Vet Admin , 11: 169, 1934
24 BAKER, J P Sickle cell anemia Virginia Med Monthly, 69: 208, 1942
25 BARANDES, R D AND KANE, T J Sickle cell anemia; sickle cell accelerating factor and its relationship to lysozyme Mil Surg , 103: 271, 1948
26 BARR, D Discussion of Ball, R P and Legant, O Sickle cell anemia in adults roentgenographic findings Radiology, 51: 695, 1948
27 BAUER, J Sickle cell disease, circulatory stasis in small blood vessels. Acta Med Scandinav , 129. 1, 1947
28 BAUER, J Sickle cell disease pathogenic, clinical and therapeutic considerations Arch Surg , 41: 1344, 1940
29 BAUER, J T Siderofibrosis of the spleen secondary to sickle cell anemia, three case reports Bull Ayer Clin Lab , Pennsylvania Hosp , 3: 477, 1946.
30 BAUER, J AND FISHER, L J Sickle cell disease with special regard to its non anemic variety Arch Surg , 47: 553, 1943.
31 BEACHAM, W D AND BEACHAM, D W Sickle cell disease and pregnancy. Am J Ob & Gyn , 60: 1217, 1950
32 BECK, J S P AND HERTZ, C S : Standardizing sickle cell method and evidence of sickle cell trait Am J Clin Path , 5: 325, 1935
33 BEET, E A Sickle cell disease in Northern Rhodesia E African Med. J , 24: 212, 1947

they have an increased mechanical fragility and the viscosity of the blood is increased. The defective erythrocytes in this disease have a life span much shorter than normal and are doomed to premature destruction in the circulation. The increased hematopoiesis that occurs in response to this is inadequate to compensate for the excessive hemolysis and a hemolytic type of anemia develops. In addition to the hemolytic anemia, the disease is characterized by the occurrence of localized lesions in the various organs in the body. These appear to result from ischemic necrosis or from thrombosis associated with infarction and hemorrhage.

As a result of the chronic anemia and repeated vascular complications, various symptoms occur. Growth and development are often retarded. Because the vascular manifestations show a striking variation from patient to patient, and from time to time in the same patient, evidence of involvement of the heart, the central nervous system, the skeleton, the skin, the urinary tract or any of the intra-abdominal organs may dominate the clinical picture. At times, the illness may appear mild and chronic, while at other times, the picture of a severe, acute illness may develop quite suddenly

The disease is present at birth, remains throughout life and proves fatal in the vast majority of patients before the normal life span is reached Studies now in progress in various places will undoubtedly increase our understanding of the various aspects of this disease It is hoped that our increased knowledge will soon lead to a more effective means of treating the persons who have the misfortune to be afflicted with this disease

ACKNOWLEDGEMENT

The authors wish to thank Mrs Robert Sullivan, Miss Brooks Overton, and Dr. Lilliane Pinero for their invaluable help in the preparation of this manuscript

BIBLIOGRAPHY

1 Abbasy, A S Sickle cell anemia first case reported from Egypt Blood, 6: 55, 1951
2 Abbott, P H The sickle cell trait among the Zande tribe of southern Sudan E African Med J 27: 162, 1950
3 Abel, M S and Brown, C R Sickle cell disease with severe hematuria simulating renal neoplasm J. A M A, 136: 624, 1948

hemolytic nature. Am J M Sc, 173: 168, 1927
1927
meningitis
Ped, 3: 764,
1949.

59. BRUGSCH H. G. AND GILL, S.: Polyarthritis in sickle cell anmeia New Eng. J. Med, 231: 291, 1944.
60. BUENO TORRES, S. Sicklemia, with report of a case Bull. Assoc. Med Santiago, 2: 43, 1944
61 BUNTING, H.: Sedimentation rates of sickled and non-sickled cells from patients with sickle cell anemia Am J Med. Sci, 198: 191, 1939
62 CABRERA CALDERIN, J G, LABOURDETTE SCULL, J. M AND BARRERAS, L. Crisis dolorosa paroxisticas, a predominancia abdominal, en el curso de una anemia a hematies falciformes Arch de med inf, 11: 61, 1942.
63 CABRERA CALDERIN, J G, LABOURDETTE SCULL, J M AND PRADO VARGAS, G.: Dos casos de anemia o hematies falciformes Bol soc cubana de pediat, 9: 179, 1937
64 CACCAMO, L P. AND STRUTNER, L. A.: Sickle cell anemia simulating rheumatic fever in the white race Ohio State Med J, 47: 121, 1951
65 CALCAGNO, P. L, MCLANY, J. AND KELLY, T. Glomerular filtration rate in children with sickle cell disease Ped, 5: 127, 1950
66 CALERO, C: Drepanocytemia and sickle cell anemia on Isthmus of Panama with special reference to its clinical manifestations and evidence by race Arch Hosp Santo Tomas, 1: 27, 1946.
67 CALLENDER, S T. E. AND NICKEL, J F. Survival of transfused sickle cells in normal subjects and of normal red blood cells in patients with sickle cell anemia Jour Lab. & Clin Med, 32: 1397, 1947
68 CALLENDER, S T. E, NICKEL, J F., MOORE, C V AND POWELL, E O. Sickle cell disease studied by measuring the survival of transfused red blood cells Jour. Lab & Clin Med, 34: 90, 1949
69 CAMPBELL, E H, JR: Acute abdominal pain in sickle cell anemia Arch Surg, 31: 607, 1935
70 CANTER, H E Sickle cell anemia in pregnancy. Med. Record, 163: 99, 1950
71. CAPRIGLIANE, L: Sickle cell anemia Arg clin, 1: 9, 1945
72 CARDENAS, PAPO, M D.: Sobre una observacion de sickle-cell anemia c hiper paratiroidismo Trab d Serv endocrinol, nutricion y sist neuro-veget, 1 27, 1942
73 CARNEVALE, A: Su di caso di anemia drepanocitica in um bambino di 26 mesi con particolare reguardo alla pathogenesis delle fasi acute della malattia Pediatria, 56: 38, 1948
74 CARRERA, G M La anemia de celulas falciformes y el embarago Clinec y obst Mexico, 4: 105, 1949
75 CASTANA, I Gigantocitie le anemie semilunari Pediatrica, 33: 431, 1925
76 CASTELLANOS, FONSECA, E Hemodistrofias por hematies falciformes Rev med.-quir de Oriente, 4: 239, 1943
77 CASTELLI, G D Giant forms and semi-lunar bodies in blood of malaria patients in Somaliland relation to sickle cell anemia Gior di med mil, 83: 847, 1935
78 CAFFEY, J The skeletal changes in the chronic hemolytic anemias (erythroblastic anemia, sickle cell anemia and chronic hemolytic icterus) Am J Roentgenol, 37: 293, 1937
79 CANBY, C B, CARPENTER, G AND ELLMORE, L F Drepanocytosis and an apparently acute surgical condition of the abdomen Arch Surg, 48: 123, 1944
80 CARANGELO, J AND OTTS, O, JR Sickle cell anemia in pregnancy South Med J, 40: 1016, 1947

34. BEET, E. A.: Sickle cell disease in a Balovale District of Northern Rhodesia E. African Med. J., 23: 75, 1946
35 BEET, E. A.: Splenic abscess and sickle cell disease. E. African Med. J, 26: 180, 1949.
36. BEET, E. A : The genetics of the sickle cell trait in a Bantu tribe Ann. Eugenics, 14: 19, 1949.
37. BELL, A. J, KOTTE, R H, MITCHELL, A. G., COOLEY, T. B AND LEE, P: Sickle cell anemia, report of two cases in young children in which splenectomy was performed. Am. J. Dis Child., 34: 923, 1927.
38 BENNETT, G A. Sickle cell anemia; further investigation of case of splenic atrophy with calcium and iron incrustations (nodular splenic atrophy). Arch Path, 7: 801, 1929
39 BENNETT, G A Splenic atrophy with calcium and iron incrustations (nodular splenic atrophy). Arch Path, 7: 71, 1929.
40 BERGMAN, P. S AND BENNE, R. M.: Tuberculoma of the brain associated with sickle cell anemia. J Mt. Sinai Hosp, 16: 175, 1949.
41 BERK, L AND BULL, G M Case of sickle cell anemia in Indian woman Clin Proc., 2: 147, 1943
42. BISHOP, F W. Elliptical human erythrocytes. Arch Int. Med, 19: 383, 1914
43. BLACK, J. E Sickle cell anemia in young infants a report of two cases J. Pediat 36: 621, 1950.
44 BLOCH, H, WALDRON, R J. AND COGAN, G. M Sickle cell disease J Pediat 38: 88, 1951.
45. BOETHE, F A Splenectomy for sickle cell anemia Ann. Surg, 97: 146, 1933
46 BOSSELMAN, B AND KRAINES, S H. Mental changes including aphasia in a patient with sickle cell anemia Am J Psychiat, 94: 709, 1937
47. BOTURAO, EDGARD AND BOTURAO, EDMIR Doenca por hematias falciformes (sickle cell disease), incidencia na Santa Casa de Santos—observacoes clinicas c hematologicas Hospital, Rio de Janerio, 32: 709, 1947
48 BOURNE, A W AND WILLIAMS, L H Recent Advances in Obstetrics and Gynecology Churchill, Ltd, London, England, 1948
49 BRANCH, H E Sickle cell anemia in a six months old colored female infant J Michigan Med. Soc, 32: 35, 1933
50. BRANDAU, G M Incidence of the sickle cell trait in industrial workers Am J Med. Sci, 180: 813, 1930
51 BRANDAU, G. M. Sickle cell anemia, report of a case Arch Int Med, 50. 635, 1932
52 BRANDAU, G M Signs of endocrine disturbance (hypogonadism) in sickle cell anemia M Rec & Ann, 38: 868, 1944
53. BRANNON, E S, MERRILL, A J, WARREN, J V AND STEAD, E A, JR The cardiac output in patients with chronic anemia as measured by the technique of right atrial catheterization J Clin Invest, 24: 332, 1945
54 BRAY, W. E Personal communication
55 BRIDGERS, W H: Cerebral vascular disease accompanying sickle cell anemia Am J Path, 15: 353, 1939

58. BROWNE, E F Sickle cell anemia Med Clin N Am, 9. 1191, 1926

105 Costa, A.: Anemias de celulas falciformes Bal Inst. Puericult Rio, 6: 56, 1948.
106 Craistopol, P. and Stewart, C F.: Acute abdominal manifestations in sickle cell disease Arch. Surg, 59: 993, 1949.
107 Cummer C. L and La Rocco C. G : Two cases of chronic ulcers of legs in patients with sickle cell anemia Arch. Derm & Syph, 39: 168, 1939
108 Cummer, C. L and La Rocco, C. G.: Ulcers on the legs in sickle cell anemia. Arch Derm & Syph, 40: 459, 1939
109 Cummer, C. L and La Rocco, C. G.. Ulcers of the legs in sickle cell anemia. Arch Derm & Syph, 42: 1015, 1940.
110 Daland, G A. and Castle, W. B : A simple and rapid method for demonstrating sickling of the red blood cell The use of reducing agents. J Lab. & Clin. Med, 33: 1082, 1948
111 Dale, G C. Sickle cell anemia. South J. Med & Surg, 99: 14, 1937.
112 Dale, M : Sickle cell anemia complicated by pregnancy. J. Michigan Med Soc, 48: 1484, 1949.
113 Dameshek, W. Familial Mediterranean target-oval cell syndrome Am J. Med Sci, 205: 643, 1943
114 Danford, E A., Marr, R and Elsey, E C.. Sickle cell anemia with unusual bone changes Am J. Radiol, 45: 223, 1941.
115 Dawson, G R., Jr. Priapism: report of five cases, two cases occurring with sickle cell anemia J. Urol, 42: 821, 1939
116 Dickstein, B and Walman, I J.. Sickle cell anemia: recent progress of pediatric interest. Am J Med Sci, 213: 728, 1947.
117 Diggs, L W.: The blood picture in sickle cell anemia South Med J, 25: 615, 1932
118. Diggs, L W.: Negative results in the treatment of sickle cell anemia Am J Med Sci, 187: 521, 1934
119 Diggs, L W.: The sickle cell phenomenon: the rate of sickling in moist preparations J. Lab. & Clin. Med., 17: 913, 1932
120 Diggs, L W.. Sickle cell trait (sicklemia). The Mississippi Doctor, 25: 64, 1947.
121 Diggs. C. W.: Siderofibrosis of the spleen in sickle cell anemia J A M A., 104: 583, 1935
122 Diggs, L W, Ashmann, C F and Bibb, J : The incidence and significance of sickle cell trait Ann. Int. Med., 7: 769, 1933
123 Diggs, L W and Bibb, J. The erythrocyte in sickle cell anemia, morphology size, hemoglobin content, fragility and sedimentation rate J A M A, 112: 695, 1939
124 Diggs, L W and Ching, R E. The pathology of sickle cell anemia South Med J, 27: 839, 1934
125 Diggs, L W and Petitt, V D A comparison of methods used in the detection of the sickle cell trait. J Lab & Clin Med, 25. 1106, 1940
126 Diggs, L W, Pulliam, H N and King, J C The bone changes in sickle cell anemia South Med J, 30: 249, 1937
127 Dolgopol, V B and Stitt, R H Sickle cell phenomenon in tuberculous patients Am Rev Tuberc, 19: 454, 1929
128 Doyle, W J and Annunziato, D Erythroblastosis fetalis in a premature infant from a mother with sickle cell anemia, a report of a patient successfully treated by exchange transfusion J Pediat, 32: 203, 1948
129. Doyle, W J and Battaglia, J The diagnosis of sickle cell anemia N. Y State J. Med, 48: 907, 1948

81. CARDOZA, W. W : Immunologic studies of sickle cell anemia. Arch Int Med, 60: 623, 1937.
82. CARROLL, O S AND EVANS, J. W.: Roentgen findings in sickle cell anemia Radiology, 53: 834, 1949
83 CHAPMAN, D. W AND CLINE, D. T.· Sickle cell anemia and heart disease J. Nat Med. A, 42: 11, 1950
84 CHEDIAK, M, CABRERA, J. AND PRADO Y VARGAS, C.: Anemia a hematias falciformes. Contribucion a su estudio en Cuba. Arch. de Med Int (Cuba), 5: 313, 1939
85 CHING, R E AND DIGGS, D W Splenectomy in sickle cell anemia, report of a case with necropsy in an adult on whom splenectomy was attempted Arch Int. Med, 51: 100, 1933
86 CLARK, F Sickle cell anemia in the white race with a report of two cases Nebraska Med J, 18: 376, 1933
87. COHEN, S M, MILLER, B W. AND ORRIS, H. W Fatal sickle cell anemia in one month old infant J. Pediat, 30: 468, 1947.
88 COLE, W. H, WALTER, L AND LIMARZI, L R Indications and results of splenectomy Ann Surg, 129: 702, 1949
89 COMBY, J Anémie à cellules falciformes Arch de med d enf, 31: 489, 1928.
90 Committee Third, fourth and fifth reports of the committee for the classification of the nomenclature of cells and diseases of the blood and blood forming tissues Am J Clin Path, 20: 3, 1950
91 CONLEY, J R, MARTIN, M B AND RECINOS, A J Sickle cell anemia Clin. Proc Child Hosp Washington, D C, 6: 101, 1950
92 CONNELL, J H Cerebral necrosis in sickle cell disease J A M A, 118: 893, 1942
93 COODLEY, E L AND KERT, M J Sickle cell disease simulating advanced rheumatoid arthritis, report of a case Calif Med, 70: 459, 1949
94 COOK, J E AND MYER, J Severe anemia with remarkable elongated and sickle-shaped red blood cells and chronic leg ulcer Arch Int Med, 16: 644, 1915
95 COOK, W C Case of sickle cell anemia anemia associated with subarachnoid hemorrhage J Med, 11: 541, 1930
96 COOKE, J V AND MACK, J K Sickle cell anemia in a white American family J Pediat, 5: 601, 1934
97 COOLEY, T V Brenneman's Practice of Pediatrics (A case report) W F Prior Co, Inc, Hagerston, Md, 3: 54, 1946
98 COOLEY, T B AND LEE, P A series of cases of splenomegaly and peculiar bone changes in children with anemia Tr Am Pediat Soc, 37: 29, 1925
99 COOLEY, T B AND LEE, P Observation on the sickle cell phenomenon Tr Am Pediat Soc, 38: 58, 1926
100. COOLEY, T B AND LEE, P Sickle cell anemia in a Greek family Am J Dis Child, 38: 103, 1929
101 COOLEY, T B AND LEE, P The sickle cell phenomenon Am J Dis Child., 32: 334, 1926
102 COOLEY T. B, WITWER E R AND LEE, P Anemia in children (with splenomegaly and peculiar changes in bones) Am J Dis Child, 34: 347, 1927
103 CORNBLEET, T, SCHOOR, H C AND BARSKY, S Pseudo-ophiasis and sickle cell anemia Arch Derm & Syph, 59: 519, 1949
104 CORRIGAN, J. C AND SCHILLER T W Sickle cell anemia A report of eight cases, one with necropsy. New Eng J Med, 210: 410, 1934

156 GORDON, R S AND GARDNER, H T . Sickle cell anemia simulating poliomyelitis in a white adult, Am J Med., 10: 528, 1951
157. GRAHAM, G. S.: A case of sickle cell anemia with necropsy Arch Int Med 34: 778, 1924
158 GRAHAM, G. S. AND McCARTY, S H · Notes on sickle cell anemia J Lab & Clin Med , 12: 536, 1927.
159 GRAHAM, G S. AND McCARTY, S H . Sickle cell (meniscocytic) anemia South Med. J , 23: 598, 1930
160 GRANADAY, J T. W An investigation to determine the earliest age of sickle cell anemia or sickle cell trait in the newborn Harlem Hosp Bull , 1: 154, 1949
161 GRANICK, S · Chemistry and functioning of the mammalian erythrocyte Blood, 4: 404, 1949
162 GREEN, T W AND CONLEY, C L · Occurrence of symptoms of sickle cell disease in the absence of persistent anemia Ann Int Med , 34: 849, 1951
163 GREENWALD, L AND BURRETT, J B Sickle cell anemia in a white family Am J Med Sci , 199: 768, 1940
164 GREENWALD, L., SPIELHOLZ, J B AND LITWINS, J Sickling trait in a white adult associated with hemolytic anemia, endocarditis and malignancy Am J Med , Sci , 206: 158, 1943
165 GRINNAN, A G Roentgenologic bone changes in sickle cell and erythroblastic anemia, nine cases Am J Roentgen & Radiol Ther , 34: 297, 1935
166 GROVER, V The clinical manifestations of sickle cell anemia Ann Int Med , 26 843, 1947
167 GULLIVER, G Sickle cells in deer Edinburgh and Dublin Philosophical Mag & J Sci , 1: 325, 1940 (quoted from Margolies (275))
168 GUTHRIE, C G. AND HUCK, J G. On the existence of more than four isoagglutinin groups in human blood Bull Johns Hopkins Hosp , 34: 37, 1923
169 GUTTON, R A AND HEINLE, R W Sickle cell anemia in the white race, study of a family with a review of genetic theories Am J Med Sci , 22: 272, 1950
170 HADEN, R L AND EVANS, F D Sickle cell anemia in the white race, improvement in two cases following splenectomy Arch Int Med , 60: 133, 1937
171 HAHN, E V Sickle cell (drepanocytic) anemia, with report of second case successfully treated by splenectomy and further observations on the mechanism of sickle cell formation Am J Med Sci 175: 206, 1928
172 HAHN, E V AND GILLESPIE, E B Sickle cell anemia report of a case greatly improved by splenectomy, experimental study of sickle cell formation Arch Int Med , 39, 233, 1927
173 HALPERN, B C AND FABER, H K The cardiopathy of sickle cell anemia and its differentiation from rheumatic carditis J Pediat , 30 · 289, 1947
174 HAM, T H AND CASTLE, W B Mechanism of hemolysis in certain anemias significance of increased hypotonic fragility and of erythrostasis J Clin Invest , 19: 788, 1940
175 HAM, T H AND CASTLE, W B Relation of increased hypotonic fragility and of erythrostasis to the mechanism of hemolysis in certain anemias Trans Assoc Am Physio , 55: 128, 1940
176 HAMILTON, J F A case of sickle cell anemia U S Vet Bur Med Bull , 2: 497, 1926
177 HAMILTON, J F A case of sickle cell anemia Memphis Med J , 11: 253, 1925
178 HAMLIN, W F Sickle cell anemia with far advanced pulmonary tuberculosis Med Bull Vet Admin , 16: 82, 1939

130. Dresbach, M : Elliptical human red corpuscles. Science, 19: 469, 1904
131. Dresbach, M : Elliptical human erythrocytes (a supplementary statement). Science, 21: 473, 1905
132 Dreyfoos, M.. Sickle cell anemia. Arch. Pediat., 43: 436, 1926
133 Eastland, J. S and Higgins, I B.· Sickle cell anemia Bull School Med, Univ. Md, 14: 178, 1930
134 Ebert, M H Sickle cell anemia with ulcer of the left ankle Arch Derm & Syph., 44: 918, 1941.
135 Editorial. Sickle cell anemia, a race specific disease. J A. M. A, 133: 33, 1947.
136 Egeli, E. S and Ergun, S.. Sickle cell anemia in a white patient. Quat tip cem mec, 12: 251, 1946
137 Emmel, V · A study of the erythrocytes in a case of severe anemia with elongated and sickle shaped red blood cells Arch Int Med, 20: 586, 1917.
138 English, R B Sicklemia occurring in Africans in Northern Rhodesia S. African Med J, 19: 431, 1945
139 Evans, R W. Sickling phenomenon in the blood of West African natives Tr Roy Soc Trop Med & Hyg, 37: 281, 1944.
140 Evans, R W. Anemia associated with the sickle cell trait in British West African natives Tr. Roy. Soc Trop Med. & Hyg, 39: 207, 1945
141. Evans, W.. Elliptical erythrocytes J Path. & Bact, 55: 378, 1943
142 Feingold, B F and Case, J T Roentgenologic skull changes in anemias of childhood Report of a case, a few notes on similar findings among skulls of Peruvian Indians Am. J Roentgenol, 29. 194, 1933
143 Felter, F and Schnabel, T G Sickle cell anemia in patients over 45, report of two cases Geriatrics, 4: 390, 1949
144 Fenichel, R L, Watson, J and Eirich, F Electrophoretic studies of the plasma and serum proteins in sickle cell anemia. J. Clin Invest, 29 1620, 1950
145 Fetter, W J. A case of sickle cell anemia complicating pregnancy Bull School Med, U Md, 32: 210, 1948
146 Findlay, G M, Robertson, W M and Zacharias, F J Incidence of sickling in West Africa Tr Roy Soc Trop Med & Hyg, 40: 83, 1946
147 Fineberg, S K and Eisenbud, K.· Sickle cell anemia with typhoid fever and multiple complications, N Y State J Med, 48: 1041, 1948
148. Flor Jorganes, J Anemia a hematies falciformes Medica, Matanzas, 2: 107, 1943
149 Forbes, G E Sickle cell anemia complicated by valvular heart disease N Carolina Med J, 10: 261, 1949
150. Ford, F R. Diseases of the Nervous System in Infancy, Childhood and Adolescence, p 642 Springfield, Ill, Charles C Thomas, 1937
151 Fouche, H H and Switzer, P K Pregnancy with sickle cell anemia, review of literature and report of cases Am J Ob & Gyn, 58: 468, 1949
152 Fradkin, W Z and Schwartz, L S Sickle cell anemia J Lab & Clin Med, 15: 519, 1930
153 Frazier, C A and Rice, C E Neonatal sickle cell anemia J A M A, 143. 1065, 1950
154 Getzoff, P. C Priapism and sickle cell anemia, a report of three cases J Urol, 48 407, 1942
155 Goodwin, W E, Alston, E F and Semans, J H Hematuria and sickle cell disease unexplained gross, unilateral renal hematuria in Negroes, coincident with the blood sickling trait J Urol, 63· 79, 1950

203 HUGHES, J G, DIGGS, L W AND GILLESPIE, C E.. The involvement of the nervous system in sickle cell anemia. J. Pediat, 17: 166, 1940
204 HUNTER, O B.: Discussion of potassium sulphocyanate a note on its use for the painful crisis in sickle cell anemia Tr Am Therap Soc., 31: 196, 1931
205. HUNTER, W. C A further study of a white family showing elliptical erythrocytes. Ann Int. Med, 6: 775, 1932
206 HUNTER, W C AND ADAMS, R B. Hematologic study of three generations of a white family showing elliptical erythrocytes Ann Int. Med, 2: 1162, 1929
207 ILLINGWORTH, C. F. W.: Formation of gall stones Edinburgh Med. J, 43: 481, 1936
208 INGBAR, S H AND KASS, E H.: Sulfhydryl content of normal hemoglobin and hemoglobin in sickle cell anemia Proc Soc Exp Biol & Med, 77: 74, 1951
209 ISAACS, R.. Sickling a property of all red blood cells Science, 112: 716, 1950.
210 ITANO, H A. AND PAULING, L: A rapid diagnostic test for sickle cell anemia *Blood*, 4: 66, 1949.
211. JAFFE, R H: Die sichelzellenanaemie Virchow's Arch Path. Anat, 265: 452, 1927.
212 JAMISON, S C.: Sickle cell anemia, a case report. South Med J., 18: 795, 1925
213 JAMISON, S C.. Sickle cell anemia (report of a case) New Orleans Med & Surg J., 76: 378, 1924
214 JAMRA, M, FERREIRA, L. M. AND BOTTURA, C.. Anemia falciforme. consideracoes em torno de um caso Hosp Rio de Janeiro, 26: 207, 1944.
215 JOHNSON, F B AND TOWNSEND, E W: Sickle cell anemia; a report of thirty cases South Med & Surg, 99: 377, 1937.
216 JONES, H L, WETZELL, F E. AND BLACK, B K.: Sickle cell anemia with striking electrocardiographic abnormalities and other unusual features with autopsy Ann. Int Med, 29: 928, 1948.
217. JOSEPHS, H. W: Clinical aspects of sickle cell anemia Bull Johns Hopkins Hosp, 43: 397, 1928.
218 JOSEPHS, H. W The presence of an anti-hemolytic factor in human plasma Bull Johns Hopkins Hosp, 62: 53, 1938
219 JOSEPHS, H W. Sickle cell anemia Bull Johns Hopkins Hosp, 40: 77, 1927.
220 JOSEPHS, H W.: Studies in hemolytic anemia I Hemolysis, compensatory regeneration and erythroblastosis Bull Johns Hopkins Hosp, 62: 25, 1938
221 JOSEPHS, H W AND WINOCUR, P A pig's plasma as a source of anti hemolytic factor. Bull Johns Hopkins Hosp, 62: 70, 1938
222 JOSEY, A I Sickle cell anemia with cerebral thrombosis South Med J, 32: 915, 1939
223 KAMPMEIER, R H: Sickle cell anemia as a cause of cerebrovascular disease. Arch Neurol & Psychiat, 36: 1323, 1936
224 KAPLAN, E AND LEWIS, S R.: The effect of human plasma transfusion on the fecal urobilinogen excretion of sickle cell anemia Blood, 4: 947, 1949
224a KAPLAN, E, ZUELZER, W. W AND NEEL, J V A new inherited abnormality of hemoglobin and its interaction with sickle cell anemia hemoglobin Blood, 6: 1240, 1951
225 KASS, E H, INGBAR, S H, HARRIS, J W. AND LEY, A B Chemical abnormalities in the erythrocytes in sickle cell anemia, their relationship to sulfhydryl metabolism and the effects of ACTH Proc Am Soc Clin. Invest, J. Clin Invest, 30: 652, 1951.
226 KATO, K AND CARDOZO, C Hodgkin's disease with terminal eosinophilia occurring in a Negro child with sicklemia J. Pediat, 12: 165, 1938.

179. Hamman, L. Clinicopathological conference: a case of severe anemia with cardiac manifestations. South Med. J, 26: 665, 1933.
180. Hanno, H. A and Margolies, M. P.: Rapid carbon dioxide test for sickling Science, 112: 109, 1950.
181 Hansen-Pruss, O C: Experimental studies of the sickling of red blood cells J Lab. & Clin Med, 22: 311, 1936
182 Hardin, A S., Jr. Sickle cell anemia: changes in the vessels and in the bones. Am J. Dis Child, 54: 1045, 1937
183 Hargrove, M. D and Matthews, W R: A fatal case of sickle cell anemia with autopsy findings J Lab & Clin. Med, 19: 126, 1933
184. Harris, J W Studies on the destruction of red blood cells. VIII Sickle cell disease molecular orientation in solutions of sickle cell hemoglobin and its significance U S Atomic Energy Commission, Oak Ridge, Tenn Thorndike Memorial Lab Boston City Hospital, Harvard Med. School, p 1-5, AEOU 885, Nov 7, 1950
184a. Harris, J. W Studies on the destruction of red blood cells VIII Molecular orientation in sickle cell hemoglobin solutions. Proc. Soc Exp. Biol & Med, 75: 197, 1950
185 Harrison, F G Priapism. Penn Med J, 50: 1074, 1947
186 Harvier, P, Brumpt, L et al Anémie à drépanocytes (hématies falciformes) et ulcers de jambe Bull Acad Nat Méd, 133: 164, 1949
187 Hayem, G: Du sang et de ses alterations anatomiques, p 336 G Masson, Paris, France, 1889
188 Heilbrun, N Sickle cell anemia with autopsy Arch Path, 16: 156, 1933
189 Hein, G E, McCalla, R L and Thorne, G W. Sickle cell anemia with report of case with autopsy Am J Med Sci, 173: 763, 1927
190 Henderson, A B Sickle cell anemia Clinical study of fifty-four cases Am J. Med, 9: 757, 1950
191 Henderson, A B and Thornell, H E Observations on the effect of lowered oxygen tension on sicklemia and sickle cell anemia among military flying personnel. J Lab & Clin Med, 31: 769, 1946
192 Henderson, A B Sickle cell disease studies "in vivo" sickling and the effect of certain pharmacological agents Am J Med. Sci., 221: 628, 1951
193 Henkin, W A Collapse of the vertebral bodies in sickle cell anemia. Am J Roentgenol, 62: 395, 1949
194 Hernandez, A R and Martinez, S U First case in region of Santa Clara Arch Soc Estud Clin Habana, 33: 683, 1939
195 Herrick, J B Peculiar elongated and sickle-shaped red blood corpuscles in a case of severe anemia Arch Int Med, 6: 517, 1910
196 Higgins, W H, Jr The heart in sickle cell anemia South Med J, 42: 39, 1949
197 Higgins, W H, Jr and Toone, E C The variable clinical manifestations of sickle cell anemia Va Med Month, 76: 400, 1949
198 Hill, F S, Hughes, J G and Davis, B C Electroencephalographic findings in sickle cell anemia Pediat, 6: 277, 1950
199 Hodges, J H The effect of racial mixtures upon erythrocytic sickling Blood, 5: 804, 1950
200. Hodges, J. H. and Bernstine, J B Sickle cell anemia and pregnancy Am J Ob & Gyn, 54: 108, 1947
201 Huck, J G Sickle cell anemia Bull Johns Hopkins Hosp, 34: 335, 1923
202 Huck, J. G and Bigalow, R M: Poikilocytosis in otherwise normal blood (elliptical human erythrocytes) Bull Johns Hopkins Hosp, 34: 390, 1923

251. LEVY, M. Sickle cell anemia; a case Am J. Dis Child, 29: 669, 1930.
252 LEWALD, L T: Roentgen evidence of osseous manifestations in sickle-cell (drepanocytic) anemia and in Mediterranean (erythroblastic) anemia. Radiology, 18: 792, 1932
253 LEWIS, A W., JR: Sickle cell anemia with pregnancy Am J. Ob. & Gyn, 33: 667, 1937
254 LIPSELT, P. J. AND DICKSTEIN, L. Pregnancy and sickle cell anemia: a case report. West. J. Surg., 58: 110, 1950
255 LONDON, I M, SHEMIN, D. AND RITTENBERG, D.: The in vitro synthesis of heme in the human red blood cell of sickle cell anemia J Biol & Chem, 173: 797, 1948
256 LOWE, R C. AND ADAMS, C C.: Studies on the pathophysiology of sickle cell disease Ann. Int. Med, 22: 192, 1945
257 LUDKE, H. AND SCHULLER, L.: Ueber die wirkung experimenteller anamien auf die herfgrosse Deutsches Arch. f klin Med, 101: 512, 1910
258 MACILWAINE, W. A AND LEAVELL, B S · Personal observation
259. McCLELLAN, R. H AND ENTWISLE, R. M.: Sickle cell anemia J Lab. & Clin. Med, 19: 507, 1934.
260 McCORD, W. M, SWITZER, P K, KELLY, W. H AND CULP, F. B.: Viscosity studies of erythrocytes from persons with sickle cell disease Proc. Soc. Exper. Biol. & Med., 69: 19, 1948
261. McCORD, W. M AND MOSELY, V.: Developments in the study of sickle cell anemia J S C Med Assoc, 46: 183, 1950
262 McCORD, W. M AND MOSELY, V: Sickle cell anemia, blood viscosity and sodium tetrathionate. Proc Soc. Exp Biol. & Med, 74: 1, 1950
263. McGAVACK, T. H. AND GERMAN, W M: Sickle cell anemia in the black Carib Indians Am. J. Med Sci, 208: 350, 1944.
264 McGAVACK, T. H AND NUSSBAUM, C C.: Skin manifestations of sickle cell anemia Urol. & Cutan Rev., 46: 194, 1942
265 McKENDRY, J B R.: Sickle cell anemia associated with cardiac failure. Canad M A J, 51: 343, 1944
266 McSWEENEY, J E. J, MERMANN, A. C AND WAGLEY, P F.: Cold hemagglutinins in sickle cell anemia Am. J Med Sci, 214: 542, 1947
267. MACHADO, ORTIZ. Splenohepatic syndrome associated with sickle cell anemia and jaundice differential diagnosis, clinical, hematologic and roentgen study; case Arch Med Inf, 1: 438, 1932
268 MACHT, S H. AND ROMAN, P. W The radiologic changes in sickle-cell anemia Radiology, 51: 697, 1948
269 MAIA DE MENDONCA, J · Meniscocytemia-sua freguencia no Brasil, primeiros resultados calcados em 1,045 pesquisas, Brasil-med, 56: 382, 1942
270 MAIA DE MENDONCA, J Meniscocitemia, sua frequencia no Brasil; resultados finais encontrados em 1,974 pesquisas Arg. Inst. biol. Exercito, 5: 83, 1944
271 MAIER, R R. AND KLEIN, M Sickle cell anemia complicated by pregnancy West J Surg, 58: 224, 1950
272 MAKRYCOSTAS, K · Uber die sickelzellanamie, wein Arch f inn Med, 33: 220, 1940
273 MALLORY, T. B · Sickle cell anemia (Cabot case 27421) New Eng. J Med, 225: 626, 1941
274 MARGOLIES, M. P.: The incidence of sickling Am J. Med Sci, 221: 270, 1951
275 MARGOLIES, M P · Sickle cell anemia Medicine, 30: 357, 1951

227 Kemp, N F and Hosay, J. J . Priapism due to sickle cell anemia, report of a case Urol and Cutan Rev , 54: 67, 1950
228 Killingsworth, W P and Wallace, S A : Sicklemia in the southwest South Med J , 29: 941, 1936
229. Kimmelstiel, P Vascular occlusion and ischemic infarction in sickle cell disease Am J Med Sci., 216: 11, 1948.
230 King, A D . Sickle cell anemia Arch. Dermat. & Syph , 33: 756, 1936
231 King, J. T , Jr and Janeway, C. A . Sickle cell anemia with cardiac complications Internat Clin , 3: 41, 1937
232 Kliger, M The relationship of trauma to sickle cell anemia; report of a case Arch Pediat , 67: 82, 1950
233 Klinefelter, H F · The heart in sickle cell anemia Am. J Med. Sci , 203. 34, 1942
234 Knode, K T and Giordano, A. S. Sickle cell anemia, case (in Negro child) report J Indiana Med Assoc , 33: 78, 1940.
235 Kobak, A. J , Stein, P J. and Daro, A F.: Sickle cell anemia in pregnancy, a review of the literature and report of six cases Am J Ob & Gyn , 41: 811, 1941.
236 Korb, J H and Miyamoto, K Meniscocytosis (latent sickle cell anemia), its incidence in St Louis South Med J , 20: 912, 1927
237 Kraft, E and Bertel, G Sickle cell anemia, case report with unusual roentgen findings Am J Roentgenol , 57: 224, 1947
238 Krugh, F J Sickle cell anemia ulcer on the leg Arch Dermat & Syph , 40: 656, 1939
239 Kuhns, W J Effects of intramuscular injection of BAL into a subject with the sickle cell trait Blood, 4: 1240, 1949
240 Landon, J F and Lyman, A V Sickle cell anemia with a case report of splenectomy Am J Med Sci , 178: 223, 1929
241 Landon, J F and Patterson, H A An evaluation of splenectomy in the treatment of sickle cell anemia, the late results of two cases so treated with a summary of the present condition of all reported splenectomized patients J Pediat , 7: 472, 1935
242 Lash, A F Sickle cell anemia in pregnancy Am J Ob & Gyn , 27: 79, 1934
243 Lawrence, J S Elliptical and sickle-shaped erythrocytes in the circulating blood of white persons J Clin Invest , 5: 31, 1927
244 Lawrence, J S Human elliptical erythrocytes Am J Med Sci , 181: 240, 1931
245 Leavell, B S and MacIlwaine, W A Sickle cell anemia observations following the production of a normal erythrocyte count by transfusion Tr Am Clin & Clim Assoc , 62: 141, 1950
246 Leavell, B S , Crockett, C L and Shotten, D Unpublished data
247 Legant, O and Ball, R P Sickle cell anemia in adults, roentgenographic findings Radiology, 51: 665, 1948
247a Lehman, H and Raper, A B Distribution of the sickle cell trait in Uganda, and its ethnological significance Nature, 164: 494, 1949
248 Leivy, F E and Schnabel, T G Abdominal crisis in sickle cell anemia Am J Med Sci , 183: 381, 1932
249 Levant, B and Stept, R Priapism due to sickle cell anemia J Urol , 59: 328, 1948
250 Levy, J Sicklemia Ann Int Med , 3. 47, 1929

302 NETHERTON, E. W Sickle cell anemia with anemia and ulcer of the leg Arch Dermat & Syph, 34: 158, 1936

303 NEUDA, P M The sickle cell disease of Negroes; its significance for the bloods of other peoples. Med. Rec, 163: 101, 1950.

304 NEUDA, P M AND ROSEN, M. S Rapid method for diagnosing sickle cell disease, preliminary report J. Lab & Clin Med, 30: 456, 1945

305 NEUDA, P. M Red blood cell sensitivity in Caucasians Ann. Int Med, 31: 1024, 1949

306 NEUDA, P M.: Red blood cell sensitivity to the blood group enzyme. Science, 106: 296, 1947.

307 NOYES, R. W. Sickle cell anemia. report of a case with autopsy. Am. J Ob. & Gyn, 52: 469, 1946

308 OBERNDORF, C P Priapism of psychogenic origin Arch Neur & Psych., 31: 1292, 1934

309 OGDEN, M A Sickle cell anemia in the white race, with report of cases in two families Arch. Int Med, 71: 164, 1943

310 OHRENSTEIN, I R Incidence of sickle cell anemia with rheumatic heart disease J. Pediat., 33: 186, 1948.

311. OLIVER, W. W Staining of the processes (flagella) of human erythrocytes J. Infect. Dis, 55: 266, 1934

312 ORTIZ, A.: Anemia meniscocitica (sickle-cell-anemia). Bol Asoc med. de Puerto Rico, 32: 273, 1940

313 ORTIZ, MACHADO AND AROSTEQUI DELA O: Sobre un caso de anemia sickle cell. Arch de med inf, 10: 91, 1941

314 PAGE, E W. AND SILTON, M Z Pregnancy complicated by sickle cell anemia Am J Ob & Gyn, 37: 53, 1939

315 PARENT, J.. Sickle cell anemia. Ann soc Belge Med. trop, Bruxelles, 30: 47, 1950.

316 PATTERSON, R H, WILSON, H AND DIGGS, L W · Sickle cell anemia a surgical problem; further observations on the surgical implications of sickle cell anemia Surgery, 28: 393, 1950

317 PAULING, L, ITANO, H A, SINGER, S J AND WELLS, I C · Sickle cell anemia; a molecular disease Science, 110: 543, 1949

318 PAULING, L, ITANO, H. A, WELLS, I C, SCHROEDER, W A, KAY, L, SINGER, S J AND COREY, R B · Sickle cell anemia hemoglobin Science, 111: 459, 1950

319 PAWAN, J L Case of sickle cell anemia in Trinidad Ann Trop Med, 31: 271, 1937

320 PENBERTHY, G C AND COOLEY, T B. Results of splenectomy in childhood Ann Surg, 102: 645, 1935

321 PENNA DE AZEVEDO, A Sobre o diagnostico histologico da anemia drepanocytica Mem Inst Oswaldo Cruz, 32: 517, 1937

322 PERR, H M The laboratory diagnosis of sickle cell anemia with special reference to the diagnostic parameter Blood, 4: 179, 1949

323 PERUTZ, M F AND MITCHISON, J M: State of haemoglobin in sickle cell anemia. Nature, 166: 667, 1950

324 PHILLIPS, A A Sickle cell anemia associated with priapism; case. J. Nat. Med Assoc, 36: 88, 1944

325 PIAGGIO-BLANCO, R A, PASEYRO, P AND DIGHIERO, J Anemia de celulas falciformes con manifestaciones cardiovasculares y embaraz Arch urug med, 30: 424, 1947

276. Marie, P. L : L'anémie à hématies falciformes. Presse méd 33: 678, 1925
277. Martinack, R. E : Pregnancy and sickle cell anemia Am J. Ob & Gyn , 53: 332, 1947.
278 Mason, V. R · Sickle cell anemia J A M. A., 79: 1318, 1922.
279. Mathews, W. R Pathology of sickle cell anemia New Orleans Med & Surg J, 88: 671, 1936
280. de Mattos, A G. and Silva, J de P. Consideracoes sobre um caso de anemia falciforme. Pediat prat Sao Paulo, 17: 41, 1946
281 Mayo, W. J Surgical aspects of diseases of lymphoid organs with special reference to the spleen Coll. Papers, Mayo Clin., 21: 631, 1929
282 Meaker, S P Human sterility The Williams & Wilkins Co , Baltimore, 1934
283 Mera, B . Meniscocytemia in Colombia. Rev Fac med , Bogota, 11: 669, 1943
284 Mera, B Meniscocytemia in Colombia Bol Offic san panam , 22: 680, 1943
285 Miles, W G Sickle cell anemia, report of two cases J. Fla Med Assn , 17: 374, 1931
286 Milliken, G Case of active sickle cell anemia. Med Rec & Ann., 22: 49, 1928
287 Miyamoto, K and Korb, J. H Meniscocytosis (latent sickle cell anemia): its incidence in St Louis South Med J , 20: 912, 1927
288 Moore, S The bone changes in sickle cell anemia with note on similar changes observed in skulls of ancient Mayan Indians J Mo Med Assoc., 26: 561, 1927
289 Moragues, V Anemia semilunar o de celulas falciformes Rev Med Trop y Parasit Habana, 7: 90, 1941.
290 Morrison, M , Samwick, A A and Landsberg, E.· Sickle cell anemia in the white race, report of two cases with diagnosis by splenic puncture Am J Dis Child , 64: 881, 1942
291 Mosely, M J , Jr Sickle cell anemia as a clinical entity J Nat Med Assoc , 38· 62, 1946
292 Moser, R A and Shaw, W J Sickle cell anemia in northern Negro J A M A , 84: 507, 1925
293 Mueller, C W , Bila, R S and Lapp, W A Sickle cell anemia and pregnancy N Y State Med J , 50: 686, 1950
294 Mulherin, W A and Houseal, R W Sickle cell anemia from a pediatric point of view Tr Sect Dis Child , A M A , p 77, 1924
295 [illegible]
296. [illegible] Int Med , 23: 376, 1945
297 Murphy, R C , Jr and Shapiro, S Sickle cell disease, observations on behavior of erythrocytes (especially dissociation of combined hemoglobin) in sickle cell disease Arch Int Med , 74: 28, 1944
298 Navarro, A and Claria Olmedo, C J Anemia drepanocitica en dos hermanos adultos negroides Dia Med , B Air , 22: 2252, 1950
299 Neal, J C , Sydenstricker, V P and Torpin, R Sickle cell anemia and [illegible] 37: 1, 1948
300 N [illegible] fer-
301 N [illegible] ence to sickle cell [illegible]
301a Neel, J V , Wells, I C and Itano, H A Familial differences in the proportion of abnormal hemoglobin present in the sickle cell trait J Clin Invest., 30: 1120, 1951

347. ROBINSON, H. A.: Sickle cell anemia; etiology with report of a case J. Michigan Med Soc, 34: 388, 1935.
348 ROBINSON, S. S AND TASKES, S Chronic leg ulcers of sickle cell anemia; report of case, with reference to recognition of the disease in California. Calif. & West Med, 64: 250, 1946.
349 ROSE, C B: Some unusual x-ray findings in skulls Radiology, 13: 508, 1929
350 ROSENFELD, G · Drepanocytic anemia; case of sickle cell anemia and case of drepanocytemia, considerations on nomenclature. Hospital, Rio de Janeiro, 25: 845, 1944.
351. ROSENFELD, S AND PINCUS, J. B · The occurrence of sicklemia in the white race Am. J Med Sci, 184: 674, 1932.
352 ROSOKOFF, J. AND BRODIE, E L: Priapism complicating sickle cell anemia; a case report J. Urol, 56: 544, 1946
353 RUSSELL, H AND TAYLOR, C. G S O.· Case of sickle cell anemia. W. African Med J, 5: 68, 1932.
354. RYAN, J. E AND FULLER, R. N.: Hemorrhagic manifestations of sickle cell disease. U. S Armed Forces Med J, 2: 623, 1951
355 RYERSON, C S. AND TERPLAN, H. L · Sickle cell anemia. two unusual cases with autopsy. Folia haemat, 53: 353, 1935
356 SAAL, J.. Sickle cell anemia with polyneuritis Bull Hosp. Joint Dis, 6: 11, 1945
357 SALAZAR Y CRUZ, S · Sobre un caso de ainhum con anemia drepanocytica Rev. Sit. Leprol. y dermat, 2: 161, 1945
358 SARMENTO, A. Contribuicao para o estudo da anemia de celulas falciformes nos negros de Angola An Instit. Med. Trop, 1: 315, 1944
359 SAWAN, E AND CAWLEY, R. D Priapism due to thrombosis in sickle cell anemia Clin Proc. Child Hosp, 3: 241, 1947.
360. SCHAFER, B F. Anemia and cholelithiasis, case Med Ann Dist. Columbia, 11: 392, 1942
361 SCHNEIDER, R G, LEVIN, W C AND HAGGARD, M E Carbonic anhydrase activity in sickle cell anemia, sickle cell trait and pernicious anemia J. Lab & Clin Med, 34: 1249, 1949
362 SCHNEIDER, R G AND LEVIN, W C Production of specific antisera against sickle cell anemia erythrocytes, antibody in sicklemia sera Proc Soc Exp Biol. & Med, 75: 110, 1950
363 SCHROEDER, W A, KAY, L M, WELLS, I C, GOERKE, C AND MOSS, J.: Amino acid composition of hemoglobins of normal Negroes and sickle cell anemics J Biol Chem, 187: 221, 1950
364 SCHWARTZ, W F: Sickle cell anemia associated with ulcers on the legs Arch Dermat & Syph, 37: 866, 1938
365 SCHWARTZ, W F. Ulcers of the leg associated with sickle cell anemia in sisters. Arch Dermat & Syph, 38: 1006, 1938
366 SCOTT, R. B, CRAWFORD, R. P AND JENKINS, M Incidence of sicklemia in the newborn Negro infant Am J Dis Child, 75: 842, 1948
367 SCRIVER, J B AND WAUGH, T R Studies on a case of sickle cell anemia Trans Am Pediat Soc, 42: 88, 1930
368 SCRIVER, J B, AND WAUGH, T. R Studies on a case of sickle cell anemia. Canad M A J, 23: 375, 1930
369 SEGURA, G, DONIN, L AND GUISTI, C L First Argentine case Dia med, 14: 1116, 1942

326. PILOT, I.: Microscopic changes in sickle cell anemia. Trans Chicago Path Soc, 12: 343, 1925
327. POLLOCK, L. H. AND DAMESHEK, W.: Elongation of red blood cells in a Jewish family. Am. J Med. Sci, 188: 822, 1934
328. PONDER, E. Red cell cytochemistry and architecture Ann. N Y. Acad Sci, 48: 579, 1947
329 PONDER, E · The sickling phenomenon and its bearing on the problem of red cell structure. J Exper Biol, 21: 77, 1945.
330 PONS, J A AND OMS, M.· Incidencia del rasgo meniscocitico (eritrocitos semilunares) en Puerto Rico; informe preliminar Bol Assoc. med. Puerto Rico, 26: 367, 1934.
331. PONTONI, L. Constitutional drepanocytic erythropathy of Herrick type with report of case in Sicilian, differentiation from constitutional hemolytic jaundice and Cooley's anemia Haematologica, 20: 657, 1939
332 POWELL, W N, RODARTE, J G AND NEEL, J. V.. The occurrence in a family of Sicilian ancestry of the traits for both sickling and thalassemia Blood, 5: 887, 1950
333 PRATT-THOMAS, H R AND SWITZER, P K Cyst of the spleen in sickle cell anemia Arch Path, 49: 159, 1950
334 PRATT-THOMAS, H R AND SWITZER, P K Sicklemia; its pathological and clinical significance South Med J, 42: 376, 1949.
335 RAMON GUERRA, A U, GIANELLI, C, RIVERO SERRA, I AND GHERADI, J La anemia de celulas falciformes Arch de Pediat d Uruguay, 14: 76, 1943.
336 RAMGEL BALLUE, M Anemia de celulas falciformes (primeiro caso relatado no Rio Grande do Sul) Rev Med Rio Grande do Sul, 2: 244, 1946
337. RAPER, A B Sudden death in sickle cell disease E African Med J, 16: 14, 1949
338 RAY, E S AND CECH, R C Infectious mononucleosis in the Negro, report of three cases with one complicated by sickle cell anemia South Med J, 37: 543, 1944
339 REINHARD, E H, MOORE, C V, DUBACH, R AND WADE, L J Depressant effects of high concentrations of inspired oxygen on erythrocytogenesis, observations on patients with sickle cell anemia with a description of the observed toxic manifestations of oxygen J Clin Invest, 23: 682, 1944
340 REINHARD, E H, MOORE, C V, DUBACH, R AND WADE, L J Effect of breathing 80 to 100 per cent oxygen on the erythrocyte equilibrium in patients with sickle cell anemia J A M A, 121: 1245, 1943
341 RICH, A R The splenic lesion in sickle cell anemia Bull Johns Hopkins Hosp, 43: 398, 1928
342 RICHTER, O, MEYER, A E AND BENNET, J P · The relation of the anemia of pregnancy *to hydremia and its treatment with aqueous liver extract* and [illegible] 28: 543, 1934
[illegible] pregnancy in sickle cell anemia South
[illegible]
[illegible] rome cardiovascular en un caso de sickle-cell anemia Medica, Matanzas, 1: 223, 1942
345 ROBERTSON, W M. AND FINDLAY, G M Sickle cell anemia in West Africa Trans. Roy. Soc. Trop Med & Hyg, 40: 435, 1947
346 ROBINSON, H. A.· Sickle cell anemia bone marrow studies J Michigan Med Soc, 26: 964, 1937.

391 SINGER, K AND ROBIN, S.: Rapid test for the demonstration of sickle cells and its clinical significance. J A M. A , 136: 1021, 1948
392 SINGER, K, ROBIN, S, KING, J C. AND JEFFERSON, R N.: The life span of the sickle cell and the pathogenesis of sickle cell anemia J. Lab. & Clin Med, 33: 975, 1948
393 SKOOG, A L: Cerebral complications in sickle cell anemia South Med. J, 33: 714, 1940
394 SKOOG, A L: Sickle cell anemia with brain involvement Dis Ner. System, 6: 276, 1945
395 SMITH, E C: Postmortem report on a case of sickle cell anemia Tr Roy Soc. Trop Med & Hyg, 28: 209, 1934.
396 SMITH, E C Sickle cell anemia; report on three cases diagnosed from microscopic sections of spleen Tr. Roy. Soc Trop Med. & Hyg, 27: 201, 1933
397. SMITH, E. M, JR. AND LEWE, I A: Sickle cell anemia associated with chronic ulcers of the leg and hyperkeratotic plaques Arch Dermat & Syph, 44: 946, 1941
398. SMITH, J H, JR: Sickle cell anemia Med Clin North Am, 11: 1171, 1928
399 SNOW, A: Some laboratory findings in sickle cell anemia Am. J. Med. Technol, 3: 177, 1937
400 SNYDER, L H, RUSSELL, H AND GRAHAM, E N: Linkage between genes for sickle cells and the M-N blood types Science, 106: 347, 1947
401 SODERMAN, W. A. AND BURCH, G E Pregnancy in active sickle cell anemia. New Orleans Med & Surg J, 90: 156, 1937
402 SPECK, G, STEVENS, C W AND HALTER, P E: Sickle cell anemia and pregnancy Va Med. Monthly, 47: 183, 1950.
403. SPILZINGER, J. M: Ulcera de las piernas en la "sickle cell anemia" (anemia falcicelular) Semana med, 2: 13, 1943
404 SPIVACK, M: Sickle cell anemia complicated by pregnancy Am J. Ob & Gyn., 50: 442, 1945
405 STASNEY, J: Erythrophagocytosis and hemosiderosis in the liver and spleen in sickle cell disease Am J Path, 19: 225, 1943
406 STEINBERG, B Sickle cell anemia Arch Path., 9: 876, 1930
407 STEINFIELD, E AND KLANDER, J V Sickle cell anemia M Clin North Am, 10: 1561, 1927
408 STEVENSON, I P Sickle cell anemia Arizona Med, 3: 161, 1946
409 STEWART, W Sickle cell disease J Pediat, 35: 255, 1949
410 STEWART, W B Sickle cell anemia, case with splenectomy Am J Dis Child, 34: 72, 1927
411 SUCHETT-KAYE, A I Syphilis, tuberculosis and sickle cell anemia, report of a case Brit J Ven Dis, 24: 148, 1948
412. SULLIVAN, B H Danger of airplane flight to persons with sicklemia Ann Int Med, 32: 338, 1950
413 SWITZER, P K The incidence of the sickle cell trait in Negroes from the Sea Island area in South Carolina South Med J, 43: 48, 1950
414 SWITZER, P K AND FOUCHE, H H Sickle cell trait, incidence and influence in pregnant colored women Am J Med Sci, 216: 330, 1948
415 SYDENSTRICKER, V P Further observations on sickle cell anemia J A M. A, 83: 13, 1924
416 SYDENSTRICKER, V P Sickle cell anemia Med Clin. North Am, 12: 1451, 1929
417 SYDENSTRICKER, V P Sickle cell anemia South Med J, 17: 177, 1924

370 SEQURA, G., RADICE, J C., DONIN, L AND GUISTI, C. L Anatomicropathologic study of first Argentine case. Arch. Soc. argent. anat., 6: 47, 1944
371 SEQURA, G, RADICE, J C., DONIN, L AND GUISTI, C. L. Anatomicropathology of first Argentine case Rev. Asoc med. argent, 58: 731, 1944.
372. SEQURA, G, RADICE, J C, DONIN, L AND GUISTI, C. L: First antochthonous case in Argentina. Semana med (tomo cincuent. fasc. 1), 311: 1944.
373. SERGENT, E AND SERGENT, E., Sur des corps particuliers du sang des paludéens Compt. Rend. Soc Biol., 58: 51, 1905
374 SHARP, E A. AND SCHLEICHER, E. M Hematologic observations on sickle cell anemia Am J Clin Path, 16: 580, 1936
375. SHARP, E A. AND VONDER HEIDE, E C.: Eunuchoid habitus associated with *sickle cell anemia and the sickling trait. J. Clin Endocrinol*, 4: 505, 1941.
376. SHARP, E A. AND SCHLEICHER, E M Hematologic observations on anemias and leukemias; sickle cell and erythroblastic anemias. Am J Med. Technol, 3: 73, 1937.
377. SHARPEY-SCHAFER, E P. Transfusion and the anemic heart. Lancet, 2: 296, 1945
378 SHEN, S C, CASTLE, W. B AND FELMING, E M · Experimental and clinical observations on increased mechanical fragility of erythrocytes Science, 100 387, 1944
379 SHERMAN, I J The sickling phenomenon, with special reference to the differentiation of sickle cell anemia from the sickle cell trait Bull Johns Hopkins Hosp, 67: 309, 1940
380 SHOTTON, D, CROCKETT, C L AND LEAVELL, B S Splenectomy in sickle cell anemia report of a case and review of the literature Blood, 6: 365, 1951
381 SIGHTS, W P AND SIMON, S D Marked erythrocytic sickling in a white adult associated with anemia, syphilis, and malaria J. Med, 12: 177, 1931
382 DA SILVA, E M Absence of sickling phenomenon of the red blood corpuscles among Brazilian Indians Science, 107: 221, 1948
383 DA SILVA, E M Index of sicklemia Mem Inst Oswaldo Cruz, 42: 315, 1945
384 DA SILVA, E M ET AL Distribuicao de grupos sanguineos comuns, O,A,B,AB e incidencia do factor Rh e siclemia nacidade de Dugue de Caxias, Estado de Rio Hospital, Rio, 34: 649, 1948
385 SILVERSTONI, E AND BIANCO, I Richerche sull anemia drepanociticia e sulla malattia microdrepanociticia in Sicilia e in Calabria Policlinico (sez prat), 56: 501, 1949
386. SILVERSTONI, E AND BIANCO, I Association of constitutional microcytic anemia and sickle cell anemia in white woman Policlinico (sez prat), 53: 265, 1946
387 SILVERSTONI, E AND BIANCO, I Una nuova entita nosologica "la malattia microdrepanocitica" Hematologica, 29: 455, 1946
388. SINGER, K, CHERNOFF, A I, AND SINGER, L Studies on abnormal hemoglobin I. Their demonstration on sickle cell anemia and other hematologic disorders by means of alkali demonstration II Their identification by means of the method of fractional denaturation Blood, 6. 413, 1951
389. SINGER, K The pathogenesis of sickle cell anemia Am J Clin Path, 21: 858 1951
390. SI
crises. J

441. VAN DER SAR, A: Anemia con eritrocitos en forma de hoz en la gestacion Rev. Policlin (Caracas), 12: 1, 1943.
442. VAN DER SAR, A: Sickle cell disease (transl.) Docum neerl et indones morbis trop, 1: 270, 1949
443. VANHEL, E. Die sichelzellenanamie Ergebr. d inn. Med -n Kinderk, 52: 504, 1937
444. VOGT, E C AND DIAMOND, L K: Congenital anemias, roentgenologically considered. Am. J. Roentgenol, 23: 625, 1930
445. VRYONIS, G: Studies of the effect of intravenous administration of liver extract in patients with sickle cell anemia, an unusual response J Lab & Clin. Med, 26: 1470, 1941
446. WADE, L J. AND STEVENSON, L. D Necrosis of the bone marrow with fat embolism in sickle cell anemia Am J. Path, 17: 47, 1941.
447. WALKER, D. W. AND MURPHY, J P Sickle cell anemia complicated by acute rheumatic heart disease and massive cerebral hemorrhage J Pediat, 19: 28, 1941
448. WALLACE, S A. AND KILLINGSWORTH, W P: Sicklemia in the Mexican race Am J Dis Child, 50: 1208, 1935
449. WASHBURN, R. E: Peculiar elongated and sickle-shaped red blood corpuscles in a case of severe anemia Va Med. Semi-Monthly, 15: 490, 1911
450. WATSON, J: Personal communication to reference 191
451. WATSON, J., STAHMAN, A W. AND BILELLO, F. P. The significance of the paucity of sickle cells in new born Negro infants Am J Med Sci, 215: 419, 1948
452. WATSON, J: Study of sickling of young erythrocytes in sickle cell anemia Blood, 3: 465, 1948
453. WEEMS, H. S: Cholelithiasis in sickle cell anemia Ann Int. Med, 22: 182, 1945
454. WEIL, I F. AND LERNER, H H A case of long standing sickle cell anemia with marked bone changes Am J. Roentgenol, 60: 251, 1948
455. WEINER, S B. Sickle cell anemia in an Italian child, a case report J. Mt Sinai Hosp, 4: 88, 1937.
456. WELLS, I. C AND ITANO, H A. Ratio of sickle cell anemia hemoglobin to normal hemoglobin in sicklemics J Biol Chem, 188: 65, 1951
457. WERTHAM, F, MITCHELL, N AND ANGRIST, A The brain in sickle cell anemia Arch Neurol & Psychiat, 47: 752, 1942
458. WIGHT, R AND THOMPSON, H J, JR Cortical fissuring in osteomyelitis complicating sickle cell anemia Radiology, 55 553, 1950
459. WILCOX, R R: Secondary syphilis, sickling, malaria, sulphonamide and Herxheimer reactions in an African soldier Brit J Dermat, 59: 59, 1947
460. WILLIAMS, A W AND MACKAY, J P Rapid determination of the sickle cell trait, by the use of a reducing agent J Clin Path, 2: 141, 1949
461. WILLIAMS, H V. Human paleopathology with some original observations on symmetrical osteoporosis of the skull Arch Path, 7 839, 1929
462. WILSON, H, PATTERSON, R H AND DIGGS, L W Sickle cell anemia, a surgical problem Ann Surg, 131: 641, 1950
463. WINSOR, T AND BURCH, G E Diagnostic physico chemical blood tests Am M Med Sci, 207: 152, 1944
464. WINSOR, T AND BURCH, G E The electrocardiogram and cardiac state in active sickle cell anemia Am Heart J, 29: 685, 1945
465. WINSOR, T AND BURCH, G E Habitus of patients with active sickle cell anemia of long duration Arch Int Med, 76: 47, 1945

418 SYDENSTRICKER, V. P., MULHERIN, W. A. AND HOUSEAL, R. W.: Sickle cell anemia. report of two cases in children, with necropsy in one case Am. J Dis Child., 26: 132, 1923
419 TALIFERRO, W. H. AND HUCK, J G.: The inheritance of sickle cell anemia in man Genetics, 8: 594, 1923
420 TEXIXERIA, A. W. G Falciform erythrocytes in a native in Angala An Inst Med. Trop , 1: 365, 1944
421. TERRY, M C , HOLLINGSWORTH, E. W. AND EUGENIO, J : Elliptical human erythrocytes Arch. Path , 13: 193, 1932.
422. THOMAS, L. AND STETSON, C. A.: Sulfhydryl compounds and the sickling phenomenon Bull. Johns Hopkins Hosp , 83: 176, 1948
423 THOMAS, L A. Neurologic complications of sickle cell anemia: a report of a case with hemiplegia Harlem Hosp Bull. N. Y , 2: 155, 1950.
424 THOMPSON, R K., WAGNER, J A AND MACCLEOD, C. M.: Sickle cell disease, report of a case with cerebral manifestations in the absence of anemia. Ann Int Med , 29: 921, 1948
425 TIANT, F. R AND MORALES COELLO, J R · Drepanocytemia associated with recent secondary syphilis with perfect tolerance of arsenobismuth therapy Vida nueva, 48: 244, 1941
426 TOMLINSON, W J Abdominal crisis in uncomplicated anemia; a clincopathologic study of eleven cases with a suggested explanation of their cause Am. J. Med Sci , 209: 722, 1945
427 TOMLINSON, W J The incidence of sicklemia and sickle cell anemia in 3,000 Canal Zone examinations upon natives of Central America Am J Med Sci, 209: 181, 1945
428 TOMLINSON, W J Studies of sicklemia blood, with new method for its rapid diagnosis Am J Clin Path , 11: 835, 1941
429 TOMLINSON, W J Study of circulation of the spleen in sicklemia and sickle cell anemia Am J Path , 21: 877, 1945
430 TOMLINSON, W J AND JACOB, J E Studies of sickle-cell formation, normal saline, plasma, and sera with carbonic anhydrase, inhibitors J Lab & Clin Med , 30: 107, 1945
431 TOOL, C D AND NORTHRUP, R V Sickle cell anemia report of a case with mural thrombi in the heart J Okla State Med Assoc , 40: 231, 1947
432 TORRANCE, E G AND SCHNABEL, T G Potassium sulphocyanate, a note on its use for the painful crisis of sickle cell anemia Ann Int Med , 6: 782, 1932
433 TORRANCE, E G AND SCHNABEL, T G Potassium sulphocyanate (thiocyanate), note on its use for painful crisis in sickle cell anemia Tr Am Therap Soc , 31: 191, 1931
434 TRINCAO, C Mais um caso de anemia eliptocitica Lisboa med , 22: 127, 1945
435 TRINCAO, C O mielograma na anemia de celulas falciformes An Inst Med Trop , 3: 81, 1946
436 TRINCAO, C The sickle-cell trait in Saint Thomas Island An Inst Med Trop , 1: 381, 1944
437 TRINCAO, C AND ROLO, J Review of literature and report of cases in white Portuguese patients Lisboa med , 19: 471, 1942
438 TROWELL, H C Sickle cell anemia E African Med J , 22. 34, 1945
439 URTEAGA BALLON, O Importancia de la puncion esplenica en el diagnostico de la sickel cell anemia Rev Med Exper , Lima, 2: 177, 1943
440. VANCE, B. M AND FISHER, R C Sickle cell disease two cases, one presenting fat embolism as a fatal complication Arch Path , 32: 378, 1941

The Growth and Maturation of the Erythrocyte*

A Consideration of Some Mechanisms Responsible for Anemia and Correlation of the Clinical and Biochemical Response to Therapy

RICHARD W. VILTER, M.D. AND JOHN F. MUELLER, M.D.

Since the 17th century, when Swammerdam observed "ruddy globules", and Leeuwenhoek described the "small round globules" he saw in his microscope when examining human blood, the human erythrocyte has intrigued many investigators The fact that this non-nucleated disc is essential to life itself has compelled many men to explore its origin, function and final destruction. Thus through the years a clearer understanding of the pathologic-physiology of erythropoiesis has come about Since any attempt at a chronological, historical review or integration of this material would be unwarranted an effort will be made to present what is known today concerning the development and function of this unique cell Before proceeding further, our efforts in this monograph must be tempered by the statement that the final answer is yet to come concerning many of the subjects to be discussed and that there is by no means unanimity of thought.

Iron has been used in therapy for centuries and has been prescribed for the treatment of chlorosis since the time of Sydenham Only very recently, however, has there been any understanding of the direct relationship of iron to the formation of hemoglobin and oxidative enzymes and an appreciation of the relationship of iron to the quality of the erythrocyte which is formed. The relationship of other factors to the formation of hemoglobin is just beginning to be appreciated.

In 1926 Minot and Murphy (108) fed liver to patients with pernicious anemia and induced a remission in this disease A liver factor had been found which was essential for the normal maturation of megaloblasts of pernicious anemia to normal erythrocytes Since then the nature of this factor has been the subject of innumerable clinical and chemical investigations, stimulated by the profound effects which it exerted on the growth and maturation of all the cells of the blood forming organs. Extracts for parenteral use were made from liver and were found to be much more potent than the oral preparations. During the ensuing years more and

* Previously unreported observations from this laboratory which have been included in this paper have been carried out under a grant from the National Vitamin Foundation, Inc

466. WINSOR, T. AND BURCH, G E.: Habitus of patients with sickle cell anemia. Human Biol , 16: 99, 1944
467. WINSOR, T. AND BURCH, G. E · Rate of sedimentation of erythrocytes in sickle cell anemia Arch Int Med., 73: 41, 1944
468 WINSOR, T. AND BURCH, G E. Sickle cell anemia; "a great masquerader", easily recognizable with routine use of diagnostic parameter. J. A. M A , 129: 793, 1945
469. WINTROBE, M. M.· The cardiovascular system in anemia with a note on the particular abnormalities in sickle cell anemia Blood, 1: 121, 1946
470. WINTROBE, M M Clinical Hematology, Third ed Lea and Febiger, Philadelphia, Pa., 1951
471. WINTROBE, M. M AND LANDSBERG, J W.: A standardized technique for the blood sedimentation test. Am J Med Sci., 189: 102, 1935
472 WOLF, I. J : Fatal poisoning with oil of chenopodium in a Negro child with sickle cell anemia Arch Pediat , 52: 126, 1935.
473 WOLF, A E Sicklemia and sickle cell anemia in Surinam Acta leidensia scholae med trop , 14: 288, 1939.
474 WALLSTEIN, M. AND KREIDEL, K. V. Sickle cell anemia Am. J Dis Child , 36: 998, 1928
475 WOOFTER, A. C , DICK, W. S AND BIERRING, W L Sickle cell anemia in white patients with ulcers of the ankles, report of two cases Arch. Int. Med , 76. 230, 1945
476 YATER, W M AND HANSMANN, G H Sickle cell anemia· a new cause of cor pulmonale, report of two cases with numerous disseminated occlusions of small pulmonary arteries Am J Med Sci , 191: 474, 1936
477 YATER, W M AND MOLLARI, M · Pathology of sickle cell anemia; report of a case with death during abdominal crisis J A M. A., 96: 1671, 1931
478 YOUNGE, W A AND MACINTOSH, E F , JR Case report, sickle cell anemia with hemiplegia J Nat Med Assoc , 33: 130, 1941
479. ZERBINO, V VOLPE, A AND NORBIS, A Anemia a celulas falciformes Arch de pediat. d Uruguay, 13: 717, 1942
480 ZIMMERMAN, S L AND BARNETT, R Sickle cell anemia simulating coronary occlusion Ann Int Med , 21: 1045, 1944
481 ZIMBRING, J G Sickle cell anemia complicated by pregnancy N Y State Med , 46: 2314, 1946

(105, 85) but in some of the megaloblastic anemias related to pernicious anemia it was effective in much smaller doses than was folic acid (101) Apparently this was the factor rather than vitamin B_{12} which was fed to pernicious anemia patients by Minot and Murphy in 1926 when they induced the first clearly documented remissions in pernicious anemia. The whole liver which they fed could not have contained enough vitamin B_{12} to have been effective without intrinsic factor.

All of these factors influence the same biological reaction, the growth and maturation of the cells of the blood forming organ, yet there are major differences in their biologic effects In addition, folic acid appears to be necessary for the formation of the pigment portion of hemoglobin, whereas vitamin B_{12} is essential for the integrity of the central and peripheral nervous systems Both vitamin B_{12} and folic acid appear to exert profound effects on the epithelium of the gastro-enteric tract (glossitis is a sign which is shared in common by all the megaloblastic anemias). Metabolic antagonists of folic acid have an inhibiting effect on the growth of blast cells of acute leukemia, or if given in too great a dose, may inhibit temporarily or permanently the growth and maturation of all the bone marrow cells.

Substances with such profound metabolic effects have stimulated great interest and very active investigation. From these and related studies, a great deal of knowledge has been obtained regarding the chemical processes which must take place when cells of the blood forming organ divide and mature. A great deal has been learned of the pathologic physiology involved in the genesis of anemia due to defective erythrocyte formation This paper deals with the chemical and physiological processes, so far discovered, which are necessary for the formation of the normal erythrocyte and the manner in which abnormalities in these reactions may lead to anemia

THE DEVELOPMENTAL STAGES OF RED BLOOD CELL FORMATION

Perhaps one of the biggest topics of debate centers around the origin of the various blood cells, including the erythrocyte It is not in the scope of this paper to scrutinize each theory and discuss it fully There are, in the main, two schools of thought One, the monophyletic school, believes that all blood cells arise from one precursor which is totipotential and which may be the lymphocyte (91) The polyphyletic group on the other hand believes that each mature blood cell has its own primordial precursor, a "blast", and that in the case of the erythrocyte this may arise from endothelial cells lining the intersinusoidal capillaries of the bone marrow (46) There are gradations of thought between these two extreme theories (47) One of the reasons for this great discrepancy is that specialized tech-

more refined liver extracts have been prepared, under the assumption that the factor originally fed to patients with pernicious anemia by Minot and Murphy was being purified. The relationship of extrinsic factor (food factor) to intrinsic factor (stomach enzyme) originally demonstrated most ingeniously by Castle (24) in 1929, has been approached from every conceivable angle. Real progress, in a chemical sense, did not begin until folic acid was shown by Spies, Vilter, Koch, and Caldwell (155) in 1945 to induce remissions in pernicious anemia and in the same year by Darby and Jones (28) in sprue. It was soon apparent, however, that folic acid was not the liver factor because very active liver extracts could be prepared which did not contain folic acid. Furthermore, Vilter, Vilter and Spies (174); Heinle and Welch (78); and many others found that the neurologic manifestations of pernicious anemia progressed, at times at a very rapid rate, when patients with pernicious anemia were treated with folic acid. However, a pure crystalline substance had been obtained which could induce such profound chemical changes in the bone marrow that defective maturation of megaloblasts, myelocytes and megakaryocytes was corrected

Another crystalline compound, a red substance containing cobalt was obtained by Shorb (149) in 1948 as the result of the fortunate discovery of a test organism, *Lactobacillus lactis dorner* which facilitated the isolation of this substance from liver extracts This material is now known as vitamin B_{12} Smith (152) was able to isolate it from liver by clinical test in patients with pernicious anemia and by following the red color of the active material West (185) then showed that this material in microgram quantities induced remissions in pernicious anemia and Spies, Garcia-Lopez, Milanes, Lopez-Toca and Culver (160) obtained a similar result in sprue. It was usually ineffective in other types of megaloblastic anemia seemingly related to pernicious anemia, such as refractory megaloblastic anemia or "achrestic" anemia (113), pernicious anemia of pregnancy (62, 7, 171) and megaloblastic anemia of infancy (100). It was ineffective by mouth except in very large doses or when given in conjunction with the intrinsic factor of normal gastric juice (104, 170) It soon became apparent that vitamin B_{12} was the liver factor and the extrinsic (food) factor as well (5) Normal gastric juice containing intrinsic factor was required for absorption of vitamin B_{12} from the gastro-enteric tract

In 1948, another hematopoietic factor was isolated by Sauberlich and Baumann (137) from liver This substance was required for growth by an organism, *Leuconostoc citrovorum*, and accordingly, it was named the citrovorum factor It was closely related to folic acid and Nichol and Welch (116) discovered that ascorbic acid was essential for the conversion of folic acid to the citrovorum factor A synthetic factor, leucovorin, induced hematologic remissions in pernicious anemia in doses equivalent to folic acid

the best known of the erythroid precursors. The basophilic cytoplasm is now replaced by the pink hue of hemoglobin. All graduations between blue, gray and pink are seen. The nucleus now is small, pyknotic and deeply stained. The chromatin is in clumps and no nucleoli are present. Presumably this cell undergoes mitosis and the nucleus gradually degenerates There is a difference of opinion as to the fate of the nucleus (33). The better evidence summarized by Howell in 1890 (83) and Dacie and White in 1949 (27) seems to favor the theory that the nucleus is finally extruded and that each normoblast, therefore, gives rise to one erythrocyte However, Clemmesen, Espersen and Plum in 1948 (25), studying bone marrow tissue cultures, have reported the observation of a mature erythrocyte budding from the cytoplasm of a normoblast. Thus even the final stage of erythrocyte development is undecided (10).

Also undecided is the significance of basophilia in the erythrocyte. This staining property may be divided into three main categories, namely, polychromasia, basophilic stippling and reticulum. It has been shown beyond reasonable doubt that the basophilia in these cells is due to remnants of ribonucleic acid present in the cytoplasm of their precursors, thus, the affinity for basic dyes (33, 165). There is further agreement that increased numbers of erythrocytes showing each of these types of basophilia are seen in any regenerative process of the bone marrow such as follows hemolysis, blood loss, and response to specific therapy. For these reasons it has been assumed that these conditions represent a stage of development between the normoblast and the mature erythrocyte. If this be true then one immediately asks, does each cell go through the three forms of basophilia and if not, what is the difference? Let us examine for a moment what we know about each of these forms of basophilia

The reticulocyte is perhaps the best known of the basophilic cells. It is present in appreciable numbers in the blood of normal persons as opposed to the infrequent occurrence of the other two types of basophilia Reticulocytes are invisible in wet preparations as well as in the usual dried film stained with Wright's stain and, to be recognized, must be stained with a "vital" dye such as brilliant cresyl blue. Normally 0 5 to 1 5 per cent of the red cells are those "skein cells", as Ehrlich referred to them. These figures compare remarkably well with the calculated daily destruction of approximately one per cent of the total circulating red cell mass (58) These cells increase remarkably in the peripheral blood as a response to or reflection of increased erythropoiesis by the bone marrow from whatever cause. Under extreme conditions they may account for as high as 70 to 80 per cent of all the circulating red cells, as we have recently seen in a case of acquired hemolytic anemia Reticulocyte incubation studies both *in vitro* and *in vivo* tend to support the contention that the life span of these cells is two to

niques have been used by various investigators which have resulted in different interpretations. For example, the results of supra-vital staining technique in one laboratory may lead to entirely different conclusions than are reached by another laboratory where some other staining technique is used. At the present time it seems wise to keep an open mind concerning the origin of the erythrocyte until more definitive proof for one theory or the other is presented.

From the primordial "stem" cell almost all hematologists recognize an orderly maturation of the erythrocyte However, here terminology introduces difficulties because each writer has a particular name for a particular cell with the result that each cell has three or four synonyms. No attempt will be made to defend any one terminology If one subscribes to the view first advanced by Ehrlich, that the megaloblast is an abnormal cell type peculiar only to erythrocyte maturation factor deficiency states, then one set of names would be used. On the other hand, if one holds that in pernicious anemia there is only a histochemical abnormality which results in a cell which appears abnormal under the microscope but otherwise does not differ from the megaloblast of normal marrow, then a different classification must be used. The latter is the feeling in our laboratory and the classification first set forth by Sabin will be used (133) Synonyms will be given in each instance.

The earliest erythrocyte precursor in the bone marrow which is readily identifiable is the *megaloblast* (pronormoblast of Ehrlich) This is a large cell 12 μ in diameter with a relatively large amount of vivid blue cytoplasm compared to other "blasts" The nucleus contains homogeneous chromatin, yet tends to clump more than usual in the young cell. Nucleoli are plentiful. These cells are rare in normal marrow.

The megaloblast, after undergoing a series of mitotic divisions, matures into an *early erythroblast* (basophilic normoblast). The differentiation between *these two cells is difficult The latter is smaller, has fewer nucleoli* and more coarsely clumped chromatin However, the basophilic cytoplasm, so characteristic of the erythroid series, is prominent and in our laboratory this cell along with the megaloblast is found in increased numbers in the bone marrow of patients afflicted with pernicious anemia Normally they comprise one to four per cent of the erythroid cells in the bone marrow

The *late erythroblast* (polychromatic normoblast), the next stage in red cell development, has lost its nucleoli, much of chromatin is clumped and the cell much smaller There may be the first tinge of hemoglobin in the otherwise blue cytoplasm This cell constitutes 26 to 29 per cent of the red cell series. Mitosis continues to occur and the next stage of erythroid maturation is imminent.

The *normoblast* (orthochromatic or acidophilic normoblast) is probably

At this point it is proper to mention several other well recognized "remnants" which appear in the erythrocyte, without inferring that these cells are necessarily young forms or precursors of the mature. These remnants take several forms and carry the names of their discoverers: 1) Howell-Jolly bodies which are generally rounded, peripherally placed and stain an intense blue, and 2) Cabot rings which are composed of the same blue material which has arranged itself in a spider web circle These structures differ from basophilia in that they are nuclear remains composed of desoxyribonucleic acid and are Feulgen positive (161) However, Schleicher in 1942 (139) was able to produce structures resembling Cabot rings *in vitro* and concluded they were laboratory artefact. The significance of these forms is not fully understood. They are seen infrequently and when present are associated with either a refractory anemia, hemolytic or otherwise, or an anemia from toxic agents.

A discussion of inclusion bodies in the erythrocyte must include Heinz bodies and siderocytes The former are highly refractile inclusion bodies composed of denatured protein which are visible in unstained wet preparations but absent in the usual dry stained film They are definitely associated with irreversible damage to the red cell produced by some toxic agent, such as phenyl-hydrazin, and frequently methemoglobinemia may be a concomitant finding (183). Cases of hemolytic disease have been described in which Heinz bodies have been a prominent feature (Fertman and Doan, 57) Siderocytes on the other hand may be recognized on the routine dry stained film and may be confused with punctate basophilia. Prussian blue reaction demonstrates that these granules are iron in a heme-free state Case (20), who has studied these cells extensively, feels that these are "old" cells, about to die, and has demonstrated that upon extrusion of these granules from the cell, not only does the serum iron rise, but the red cell is then highly susceptible to phagocytosis These cells are increased in numbers following splenectomy for any cause, in lead poisoning, in untreated pernicious anemia, in uremia and in many other hemolytic diseases

Finally granules of unknown origin have been described which stain with neutral red in supravital preparations and black in ordinary stained films. They generally occur as single, highly refractile bodies According to Isaacs (84), they are found in 0 3 to 0 7 per cent of red cells in normal blood and represent the final stage in the maturation of the erythrocyte. This opinion has been neither proved nor disproved.

From this complicated morphologic maturation which has numerous pitfalls along its course, the erythrocyte emerges as the mature element. We will not quarrel with the term "cell" as used for the non-nucleated erythrocyte It serves several specific functions nobly, respires minimally

four days and that each reticulocyte "ripens" into a mature erythrocyte (193). Some investigators feel that this is the process which normally occurs in the bone marrow and that the expression of peripheral reticulocytosis is merely a release phenomenon associated with increased erythroid activity (77). However, reticulocytosis has been observed in this laboratory in patients with liver disease in whom evidence of blood loss, hemolysis or erythroid hyperplasia of the bone marrow was lacking Thus one might *wonder if the reticulocyte represents an abnormal intermediary product of* accelerated erythroid development whereas normally there is time for the orderly development of the erythrocyte and very few reticulocytes are made. In time of stress of increased production, however, the ribonucleic acid is unable to clear itself properly from the cell and a tremendous increase in numbers of "reticulated" cells results. This is pure hypothesis with little scientific fact to back it up

Punctate basophilia or stippling and polychromasia are closely related phenomena (33). They may be seen in the same cell and are easily recognized *with the common Romanowsky stains They are found rarely* in normal persons, although careful studies of a group of "normal" persons did reveal punctate basophilia in a significant percentage (56). However, stippling is known to be associated with heavy metal intoxication, especially lead, and has been thought to be pathologic (120). Therefore, it has been suggested that in lead poisoning the erythrocyte membrane is injured allowing the basic dye to enter the cell and stain the remaining cytoplasmic remnants. The latter are thought to be identical with those in the reticulocyte since in this instance reticulocytosis and stippling are often co-existent Recently, however, stippled erythrocytes have been produced (54) *in vitro* by incubating red blood cells with lead There was no relationship here between the production of stippling and reticulocytes Therefore, the exact etiology of punctate basophilia in lead intoxication remains unexplained.

What then is the relationship between these three examples of basophilia? Polychromasia closely parallels reticulocytosis in most instances It is possible artificially to change polychromasia into reticulation by merely allowing the blood to stand 48 hours Furthermore, by increasing the degree of fixation one is able to decrease the amount of reticulation Polychromasia can be changed into punctate basophilia by increasing the time of staining (33). This whole problem becomes very involved, but suffice it to say: 1) all three basophilic types are cytoplasmic in origin and result from remnants of ribonucleic acid 2) the type observed is partly dependent upon the degree of fixation, length of staining and the permeability of the cell membrane to the basic dyes, 3) punctate basophilia is diagnostic of heavy metal intoxication only when present in large numbers of cells

except insofar as they influence the all important synthesis of organic compounds Analyses may be obtained from papers by Dietz (42), Dietz and Steinberg (43), Ponder (124), and Granick (66). The important organic constituents of the cell protoplasm, many of which will enter prominently into future discussions of division and maturation, are as follows: proteins, of both general and specialized type, an example of the latter being hemoglobin, nucleoproteins; phospholipids, lipids and cholesterol; glucose and possibly glycogen; compounds containing SH groups like glutathione and cysteine; and the apoenzymes (highly specialized proteins) and their cofactors (coenzymes).

The proteins of the primitive erythroid cells of the blood forming organs contain all the essential amino acids and are biologically similar to proteins of the cells of other tissues. They are most important components of protoplasm and must be synthesized in large amounts when cells divide rapidly. Together with glucose and glycogen they supply energy for the cell. The apoenzymes are one type of specialized protein of the cells Together with their coenzymes, specific organic compounds of smaller molecular size of which many of the vitamins are precursors, they act as the respiratory and metabolic catalysts of the cells These enzyme systems speed up chemical reactions In addition there are phosphoproteins and incomplete proteins, particularly histones in the cells They occur for the most part in the nuclei in association with the nuclear nucleic acid

Hemoglobin is a specialized protein found in maturing cells of the erythrocyte series in increasing amounts as maturation progresses. It is a combination of heme with the protein, globin Heme is formed by coordinated linkage of ferrous iron to protoporphyrin. Studies with radioactive tracers indicate that the four pyrrol nuclei of protoporphyrin can be made by the body with great ease from such simple compounds as glycine and acetate. Folic acid is one of the catalysts which speed up this reaction. There is evidence that uroporphyrin and coproporphyrin may be chemical precursors of protoporphyrin Rare diseases characterized by the appearance of large amounts of uroporphyrin, coproporphyrin and porphobilinogen in urine and stool, the porphyrias, may be caused by inborn or congenital chemical blocks occurring in the reactions leading to the formation of protoporphyrin.

The nucleoproteins are complex substances combining protein, histone or protamine with nucleic acid Little is known of the way in which these combinations occur but much has been learned in the last few years about the nucleic acids and has been summarized by Schlenk (140) These substances are polynucleotides which contain the purine bases, adenine and guanine, the pyrimidine bases cytosine and either thymine or uracil each attached to a pentose sugar, ribose or desoxyribose by a glucoside linkage

and fulfills most of the criteria of a living cell. The biconcavity of the red corpuscle renders the measurement of the surface area difficult, but it has been estimated at 140 square microns (189). The cell "membrane" or "lining" has been described by Ponder (124) as composed of lipid palisades with masses of the fibrous protein, stromatin, running tangentially over the surface "like string wound loosely in a ball".

The nature of the interior of the red cell is poorly understood. We know it is composed of approximately 34 per cent hemoglobin and water, minerals, enzymes, and nucleoprotein remnants The manner in which hemoglobin is held is conjectural, but perhaps the best evidence indicates that it is in solution. One theory pictures it held within the meshes of a homogenous, jelly-like substance (107), while another hypothesis holds that the hemoglobin is in the form of a hydrophilic gel (94). These two ideas are based on the observation that the capsule may be severed without loss of its contents However, it may be argued that tampering with the cell produces coagulation of the interior and thus prevents extrusion. Nonetheless, the red corpuscle is a highly efficient structure, geared to do its job, flexible and elastic

In no other cell can the body's economy be so strikingly shown Without the O_2 and CO_2 carrying power of hemoglobin the activity of human beings would be reduced to 1/50 of that possible normally. Furthermore, if hemoglobin were free in the plasma rather than restricted to a cell its osmotic coefficient would be so great that body water would be unable to re-establish an equilibrium and tissue dehydration would ensue (2) In addition it is thought that hemoglobin functions more efficiently in a phosphate medium and at the hydrogen-ion concentration present in the red cell

THE CHEMICAL COMPOSITION OF THE CELLS OF THE ERYTHROID SERIES IN BLOOD AND BONE MARROW

Over the past twenty years a great deal of investigation has shed light on the chemical composition of cells and on the physiologic and chemical function of the various intracellular structures The cells of the blood forming organ have been particularly accessible to study by cytochemical staining techniques and chemical analysis, and there is a great deal of basic chemical information concerning them. Since division and maturation of these cells occur coincidentally with and probably because of biochemical changes in them, information concerning their biochemistry is essential to a discussion of division and maturation The biochemistry of the erythroid cells beginning with the primitive erythroblast or megaloblast is particularly important to future discussion However, our information concerning the chemistry of these cells is limited in many areas.

There is no reason for recounting the inorganic constituents of these cells

except insofar as they influence the all important synthesis of organic compounds. Analyses may be obtained from papers by Dietz (42), Dietz and Steinberg (43), Ponder (124), and Granick (60). The important organic constituents of the cell protoplasm, many of which will enter prominently into future discussions of division and maturation, are as follows: proteins, of both general and specialized type, an example of the latter being hemoglobin; nucleoproteins, phospholipids, lipids and cholesterol; glucose and possibly glycogen; compounds containing SH groups like glutathione and cysteine; and the apoenzymes (highly specialized proteins) and their cofactors (coenzymes).

The proteins of the primitive erythroid cells of the blood forming organs contain all the essential amino acids and are biologically similar to proteins of the cells of other tissues. They are most important components of protoplasm and must be synthesized in large amounts when cells divide rapidly. Together with glucose and glycogen they supply energy for the cell. The apoenzymes are one type of specialized protein of the cells. Together with their coenzymes, specific organic compounds of smaller molecular size of which many of the vitamins are precursors, they act as the respiratory and metabolic catalysts of the cells. These enzyme systems speed up chemical reactions. In addition there are phosphoproteins and incomplete proteins, particularly histones in the cells. They occur for the most part in the nuclei in association with the nuclear nucleic acid.

Hemoglobin is a specialized protein found in maturing cells of the erythrocyte series in increasing amounts as maturation progresses. It is a combination of heme with the protein, globin. Heme is formed by coordinated linkage of ferrous iron to protoporphyrin. Studies with radioactive tracers indicate that the four pyrrol nuclei of protoporphyrin can be made by the body with great ease from such simple compounds as glycine and acetate. Folic acid is one of the catalysts which speed up this reaction. There is evidence that uroporphyrin and coproporphyrin may be chemical precursors of protoporphyrin. Rare diseases characterized by the appearance of large amounts of uroporphyrin, coproporphyrin and porphobilinogen in urine and stool, the porphyrias, may be caused by inborn or congenital chemical blocks occurring in the reactions leading to the formation of protoporphyrin.

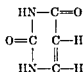
The nucleoproteins are complex substances combining protein, histone or protamine with nucleic acid. Little is known of the way in which these combinations occur but much has been learned in the last few years about the nucleic acids and has been summarized by Schlenk (140). These substances are polynucleotides which contain the purine bases, adenine and guanine, the pyrimidine bases cytosine and either thymine or uracil each attached to a pentose sugar, ribose or desoxyribose by a glucoside linkage

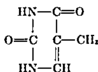
(fig. 1). Phosphate is joined to the sugar by an ester linkage at the three position. A purine, adenine, linked to ribose is called adenine riboside, adenine nucleoside or adenosine Similarly there are ribosides or desoxyribosides of the other purines and pyrimidines, guanine nucleoside or guanosine, uracil nucleoside or uridine, cytosine nucleoside or cytidine and thymine nucleoside or thymidine When these nucleosides combine with phosphate to form a phosphate ester, they are called nucleotides, such as adenine nucleotide or adenylic acid, guanine nucleotide or guanilic acid, uracil, thymine, and cytosine nucleotides or uridilic acid, thymidilic acid, and cytidilic acids. *These nucleotides are linked to each other through phos*phate by an additional ester linkage to form polynucleotides which may be polymerized to yield molecules of high molecular weight, the nucleic acids.

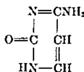
In 1920, the statement was made with confidence that there were two types of nucleic acid, one in plant nuclei characterized by the sugar, ribose, and the pyrimidine, uracil; the other in animal cell nuclei characterized by the sugar, desoxyribose, and the pyrimidine, thymine in place of uracil Thus, the terms yeast (plant) and muscle (animal or thymus) nucleic acids came into common use Since then, however, both types of nucleic acid have been found in plant and animal cells The chemical distinction, however, is still valid, and we still speak of ribose nucleic acid (yeast type) containing adenine, guanine, cytosine, uracil, ribose and phosphate and desoxyribose nucleic acid (muscle type) containing, adenine, guanine, cytosine, thymine, desoxyribose and phosphate For many years the nucleic acids were thought to be tetranucleotides, containing the purines and pyrimidines, in 1:1.1.1 ratios Recent analyses indicate that this theory is not tenable and that the purines and pyrimidines occur in different ratios in the nucleic acids from different tissues

Ribose nucleic acid is found principally in the cytoplasm of cells of the blood forming organ but it also occurs in the nucleoli of very young cells. In both sites it is responsible for the blue color of the cytoplasm and the pale blue color of the nucleoli in Wright-Giemsa stained preparations In pyronine stained preparations it is pink It is responsible for the dark blue staining cytoplasm of the erythroblasts, and the reticulum, polychromatophilia and stippling of immature erythrocytes. On the other hand, desoxyribose nucleic acid exists only in the nuclear chromatin as far as is known. It is usually combined with a histone or protamine and gives the characteristic reddish purple color to the chromatin in Wright-Giemsa stained blood or bone marrow films. In the Feulgen reaction, desoxyribose nucleic acid stains red with carbol fuchsin

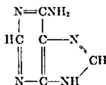
There is evidence that ribose nucleic acid influences the formation of

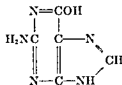

uracil


thymine (5-methyluracil)

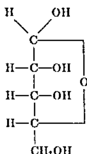

cytosine

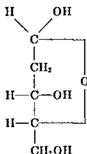
Pyrimidines


adenine

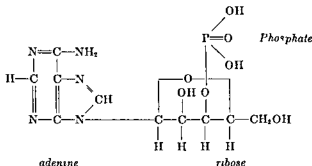

guanine

Purines


d-ribose


d-desoxyribose

Sugars



Adenosine-3-phosphate (adenine nucleotide or adenylic acid)

FIG 1 STRUCTURAL FORMULAE OF PURINES AND PYRIMIDINES OF THE NUCLEIC ACIDS AND OF A NUCLEOTIDE, ADENOSINE-3-PHOSPHATE

to a halt as cells mature, ribose nucleic acid disappears from the cytoplasm and nucleoli are no longer visible. Hemoglobin formation no longer takes place in the mature erythrocyte which is deficient in ribose nucleic acid but can continue in the reticulocyte, whose reticulum is ribose nucleic acid. Mature cells such as the lymphocyte, plasma cell and some histiocytes, one of whose functions is the continuous formation of globulins, retain their cytoplasmic basophilia (blue staining ribose nucleic acid) throughout their lives. In the molecular structure of the nucleic acids, the necessary energy for protein synthesis is available.

The desoxyribose nucleic acids of the nucleus are associated closely with the proteins of the chromosomes, histones, protamines and complete proteins. Apparently they are responsible for cell mitosis and division. It is thought that the nucleic acid molecules are attached along the polypeptide chains of these proteins and detach and reattach during mitotic division. Another protein containing tryptophane (therefore a more complete protein than histone) is also present and is associated with a small amount of ribose nucleic acid. This latter protein is supposed to give the chromosome its characteristic thread-like structure and some investigators believe it is the principal protein of the chromosomes and genes. X-ray irradiation and other nucleotoxic agents seem to interfere with the detachment of the desoxyribose nucleic acid and increase the stickiness of the chromosomes, thus interfering with division of cells. Heavy, dark staining chromatin around the nucleoli of very young cells, called the nucleolus associated chromatin (heterochromatin), controls the formation of ribose nucleic acid in the nucleolus, and by slowing this down it decreases growth activity and formation of protein in these cells as they mature. This chromatin ultimately replaces the nucleoli when maturation is complete. There is much speculation but little fact beyond this point.

Phosphorylases are necessary for the phosphorylation of the ribosides and desoxyribosides and for their ultimate esterification to form the polynucleotides. Phosphatases called nucleases, nucleotidases, and nucleosidases split the phosphate linkages and break down these complex molecules. These and many other enzymes exist in the cells of the blood forming organ and catalyze an internal metabolic cycle which splits the nucleic acids to nucleosides and then rebuilds new nucleic acids. Cytoplasmic ribose nucleic acid is undergoing constant degradation and synthesis. Nuclear desoxyribose nucleic acid is much more stable but nevertheless has a slow turn-over rate. There is some evidence that the one type of nucleic acid can be converted into the other at a slow rate. Excellent discussions on the chemistry of nucleic acids can be obtained in "Cold Springs Harbor Symposia on Quantitative Biology", M. Demerec, Editor, Nucleic Acids and Nucleoprotein (39) and in a review by Schlenk (140).

Phospholipids like lecithin and cephalin probably form a considerable portion of the supporting structure or stroma of the cells. These substances occur in cytoplasm, nucleus, mitochondria and granules of polymorphonuclear leukocytes, eosinophiles and monocytes. They are almost certainly present in the erythroblasts also Lipids are also present and increase in amount when the cell is in a noxious environment. Substances of the phospholipid class can be demonstrated cytochemically with Sudan black B and Baker's hematein test combined with pyridine extraction (9).

Glutathione, cysteine and many other substances, usually enzymes, containing free SH groups are present in the cells of the bone marrow, and are highly reactive in metabolic processes. They may be found in erythroblasts and erythrocytes. Their presence can be determined by chemical methods but their exact function in cellular respiration and metabolism other than to act as hydrogen donors or acceptors is unknown. In the leukocytes of chronic and acute lymphatic leukemia these sulphydryl compounds are increased in amount, but their concentrations in erythroblasts have not been investigated.

Enzyme-coenzyme systems may be found in all parts of the bone marrow cells Phosphatases and phosphorylases, particularly, are universally distributed (93). Other enzymes found in these cells are peptidases, carbonic anhydrase (173) (a zinc containing enzyme which splits carbonic acid) amylase, lysozyme (an antibacterial enzyme), catalase (44), cytochrome oxidase, lipase, cathepsin, lecithinase, beta-glucuronidase, oxidases, peroxidases and proteolytic enzymes. This subject has been reviewed by Rebuck (127). Many of these are found in the granules and mitochondria in high concentration. Vitamin B_{12} is found in the mitochondria particularly. Folic acid is found throughout the entire cell

There is no information concerning changes in these cellular constituents in erythroblasts but a few have been studied in normal leukocytes and those found in disease states Significant differences have been observed. This subject has been reviewed by Valentine (172) For a discussion of physical and chemical constitution of the erythrocyte see p 521 and 526

Erythrocytes are known to contain the following enzymes: catalase (44), the pyridine nucleotides (96) (nicotinamide containing coenzymes), and riboflavin adenine dinucleotide (95), carbonic anhydrase (173) and cholinesterase (134). The last is said to be a useful index of bone marrow activity In pernicious anemia it is low and rises with reticulocytosis. In most other anemias characterized by hyperactive marrow it is high.

The nucleated cells of the blood and bone marrow respire (i e. utilize oxygen) whether glucose is present in the medium or not (55) When glucose is absent respiratory quotients fall suggesting that the cells are metabolizing

fats Blast cells, including erythroblasts, have low aerobic glycolytic rates, i.e they metabolize little glucose in the presence of oxygen (180, 8). In this regard they resemble embryonic cells The mature erythrocytes on the other hand have a very low oxidative and glycolytic metabolism.

STIMULI WHICH INCREASE OR DECREASE ERYTHROID ACTIVITY

Anoxia which may occur for a variety of reasons will increase the activity of the erythropoietic tissue of the bone marrow. It will lead to an increase in the erythrocyte precursors especially the normoblasts and to their maturation into mature erythrocytes. Anoxia will induce this effect whether due to acute blood loss or hemolysis, to high altitudes and low oxygen tension in the respired air, to chronic lung disease, or to congenital heart disease with cyanosis. On the other hand, increased oxygen tension in the respired air will have the opposite effect. For instance, a patient with hemolytic anemia who breathes 100 per cent oxygen by mask for a week or ten days will no longer have a high reticulocyte count and his erythrocytes and hemoglobin may actually decrease (168). Transfusions have a similar effect in severely anemic persons whose blood forming organ has become very active to meet the need for increased numbers of erythrocytes For instance, a person with pernicious anemia and a hyperplastic megaloblastic bone marrow, after transfusions have raised his erythrocytes to a normal value, will be found to have a normoblastic and normally cellular bone marrow (30). As the anemia recurs the marrow again becomes hyperplastic and megaloblastic.

Hormones may have a similar effect upon the activity of the blood forming organ This subject has been reviewed by Gordon and Charipper (63) and Daughaday, Williams and Daland (29) In general, however, this effect is not apparent until a gland is removed and observations are made of the results of such removal and of replacement therapy Hypopituitarism, hypoadrenalism, and hypothyroidism usually result in moderate normocytic normochromic anemia and hypofunctioning bone marrow Hypophysectomized rats tend to have a microcytic hypochromic anemia and hypoplasia of the bone marrow with reticulocytopenia Thyrotropic, gonadotropic and growth hormones stimulate erythropoiesis in such hypophysectomized rats Human beings with Simmond's disease or Sheehan's syndrome usually have a mild normocytic normochromic anemia which responds to active pituitary extracts Patients with Addison's disease also tend to have a mild normocytic normochromic anemia which may be masked by the characteristic sodium depletion and dehydration The bone marrow tends to be hypoactive but responds to active adrenal cortical substances Human beings with myxedema as well as thyroidectomized rats tend to have a macrocytic anemia and hypoactive bone marrow which responds to desic-

cated thyroid or to thyroxin The effect of the gonads on hematopoiesis is indicated by the observations that male rats subjected to bleeding regenerate blood more rapidly than female rats under similar experimental conditions Testosterone increases the erythrocyte counts of castrated rats while estrogens usually depress the counts of such animals. Such evidence suggests that the sex hormones play a considerable part in maintaining the erythrocyte counts of men at a somewhat higher level than those of women. Experiences of blood banks suggest that men regenerate blood much more rapidly than women This may be because iron reserves are larger in men than women, though a sex difference may play a part also

In general, an excess of these hormones does not ordinarily increase the erythrocytes beyond normal. There are exceptions, however. Polycythemia is characteristic of patients with Cushing's syndrome due to excessive production of adrenal cortical steroids Experiences with adrenal corticotropic hormone from the pituitary and cortisone from the adrenal cortex indicate that these substances are general bone marrow activators If ACTH or cortisone is given in sufficient amounts, usually in excess of 100 mg a day for ACTH and 200 mg a day for cortisone, normal human beings will have a moderate reticulocytosis and moderate elevation of erythrocytes and hemoglobin unless these effects are masked by the water retaining action of these hormones The bone marrow usually becomes more cellular and both erythroid and myeloid activities are increased, particularly erythroid As soon as hormone therapy is discontinued marrow activity returns to its former level or may be reduced temporarily below this and erythrocytes and hemoglobin fall to pre-treatment levels

It is generally assumed that the depression of bone marrow activity observed upon removal of the pituitary, thyroid, adrenal glands or the gonads is due to a general slowing down of all metabolic processes and that recovery is due to reactivation of normal metabolism

It is possible, also, that the stimulatory effect of hypoxia, and depressant effects of hyperoxia on bone marrow activity are mediated through the pituitary gland (106, 63) In animals hypoxia fails to produce a stimulatory effect in the absence of the hypothalamus and pituitary gland.

In some unknown way the spleen regulates the activity of the bone marrow and under certain circumstances may depress one or more of its functions Hypoplasia of the erythroid cells may result in a condition which may be called "hypersplenic hormonal inhibition" Owren (118) has described such a situation at the onset of the acute hemolytic crisis of congenital hemolytic jaundice Chronic infectious diseases, inflammatory processes, renal disease, liver disease and malignant tumors may inhibit erythroid activity by mechanisms which have not yet been discovered. Drug sensitivity and x-ray irradiation may induce hypoplasia or aplasia of the

erythroid elements of the bone marrow by interfering with critical chemical reactions.

CHEMICAL PROCESSES WHICH OCCUR DURING THE GROWTH AND DEVELOPMENT OF THE ERYTHROID CELLS OF THE BONE MARROW

Many complicated chemical processes are essential for the growth and division of young erythroid cells and for their subsequent maturation into adult erythrocytes. Actively dividing erythroid cells must respire, and for this process, they are dependent upon the respiratory enzymes, many of which have B complex vitamins as precursors Deficiencies of these vitamins, particularly nicotinic acid and riboflavin, should impair the function of erythropoietic tissue. Though anemia does occur in animals as a result of deficiencies of these substances, no counterpart has been observed in man, and we cannot relate these vitamins to human blood formation as yet.

Actively dividing erythroid cells must also form new protein, particularly hemoglobin, at a rapid rate Protein formation, including globin, seems to be under control of the ribose nucleic acids of cytoplasm and nucleoli, which in turn are controlled by the nucleolar associated chromatin of the nucleus Cell division is under the control of the chromosomes, composed principally of desoxyribose nucleic acid in combination with complete proteins and histones

Therefore, two processes of great importance to the growth and maturation of erythroid cells are 1) the formation and degradation of nucleic acids and 2) the formation of hemoglobin What is known of the chemistry of these two processes will be discussed in the following sections Whenever possible, human anemias will be discussed in terms of abnormalities in these chemical reactions

The Formation of Nucleic Acids

Cytoplasmic ribose nucleic acid has a rapid turnover rate This is indicated by considerable uptake of labeled phosphorus by normal rat or rabbit liver ribose nucleic acid within hours after the administration of P_{32} (72, 31) Formation and degradation of this nucleic acid continue as long as there is a requirement for active protein formation As the protein requirements of the cell are fulfilled and maturation or aging begins ribose nucleic acid decreases and finally disappears Thus, the earliest recognizable precursor of the erythrocyte, the megaloblast or pronormoblast, has cytoplasm which is deep blue and two or more nucleoli which also are blue in Wright-Giemsa stained films, indicating the presence of much ribose nucleic acid. As the normoblast matures the cytoplasmic basophilia gradually decreases in intensity, the nucleoli disappear and increasing amounts of hemoglobin impart a polychromatophilic and finally orthochromatic

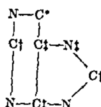
color to the cytoplasm. The greatest number of mitotic figures indicating the most active cellular division occur during the early and late erythroblast stage when protein formation is most rapid. As more and more hemoglobin appears, less and less mitotic activity is evident.

On the other hand, nuclear desoxyribose nucleic acid is relatively stable (31). Once it is formed there is only a very slow turnover rate. Changes must take place, however, in its structure and function as indicated by the morphologic changes evident under the microscope. In the megaloblast (pronormoblast) the chromatin is fine and thread-like. As the cell grows older, these threads become coarser and finally form dense aggregates of chromatin, a condition called pyknosis, and one that is characteristic of the normoblast nucleus. Ultimately this pyknotic nucleus is lost (see p. 523). When erythropoietic activity is great these newly formed non-nucleated erythrocytes still contain small amounts of ribose nucleic acid as indicated by cytoplasmic basophilia and reticulum While this material remains, hemoglobin synthesis is still possible It disappears, however, in 24 to 48 hours and thereafter the hemoglobin content of the cell is fixed for the remainder of its life span of 120 days These nucleoprotein-protein relationships have been clearly demonstrated spectrophotofluorometrically by Caspersson (23), Caspersson and Schultz (21), Caspersson and Santesson (22), Thorell (167) and others. Davidson (31) confirmed these observations by the chemical determination of the nucleic acid content of these cells at various stages of bone marrow activity.

These nucleic acids are formed from simple compounds like the amino acid, glycine, which can contribute both carbon and nitrogen atoms, and from the carbon atoms of formate and carbon dioxide. There is little evidence that they are formed to any great extent from dietary purines or pyrimidines though rats fed radioactive adenine have some radioactivity in both adenine and guanine (12) of the nucleic acids Plentl and Schoenheimer (123) showed that N_{15} labeled free pyrimidines are not utilized for nucleic acid synthesis in the rat, but their nucleosides, uridine and cytidine, are utilized Such experiments suggest that, to some extent, ingested purines may be incorporated directly into nucleic acids or may be converted to other purines before such incorporation. Pyrimidines are not usually utilized for formation of nucleic acids though their nucleosides are. Investigations in animals with glycine, formate and carbon dioxide labeled with C_{14} have shown that the purine ring usually arises *de novo* from these substances (52, 122) (fig 2) The origin of the pyrimidine ring is not so clearly defined Wright, Miller, Skeggs, Huff, Weed and Wilson (191) have suggested that aspartic and orotic acids are precursors Oxalacetic acid and carbon dioxide may also be precursors according to Mitchell and Houlahan (109) (fig 3).

One may deduce from Greenberg's work (69) that before the purine and

pyrimidine rings are closed or substituted groups or side chains are added, ribose or a ribose phosphate ester must be attached so that actually a nucleoside or nucleotide is formed when the ring is completed (fig 4).



* From CO_2. † From formate ‡ From glycine.

FIG 2 CONTRIBUTION TO THE PURINE RING BY GLYCINE, FORMATE AND CARBON DIOXIDE.

Nitrogen is obtained from many sources—called the nitrogen pool

HO—C═O | CH_2 | O═C—COOH
oxaloacetic acid

$NH_2 + CO_2$ →

HN—C═O | O═C C—H | N—C—COOH | ribose
orotic acid riboside

HN—C═O | O═C C—H | N—C—H | ribose
uracil riboside

HN═C—NH_2 | O═C C—H | N—C—H | ribose
cytosine riboside

FIG 3 [illegible] FROM OXALO- [illegible] rather than

Free purines and pyrimidines may never be formed, a possible reason for the low physiologic activity of such substances when given orally. These nucleotides are then joined through their respective phosphate groups by esterification and ultimately they are polymerized Many enzyme systems are necessary to activate these processes The action of only a few has been

established but for these folic acid and vitamin B_{12} are essential activators (148).

THE METABOLIC EFFECTS OF FOLIC ACID

In recent years evidence has accumulated which indicates that one of the principal functions of folic acid is acceleration of the transfer of single carbon units. For instance, Sakami (135), Elwyn and Sprinson (52), and many others showed that folic acid is necessary for the reversible reaction, glycine⇄serine which requires the addition or subtraction of a single carbon unit (fig. 5).

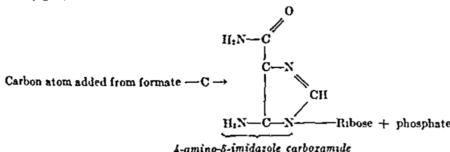


FIG. 4. FORMATION OF A PURINE NUCLEOTIDE, INOSINIC ACID, BY CLOSURE OF RING OF 4-AMINO-5-IMIDAZOLE CARBOXAMIDE AFTER THE ADDITION OF RIBOSE PHOSPHATE.

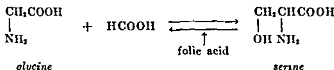


FIG 5

Folic acid is also essential for the reaction which involves the transfer of the single carbon unit of formate to homocysteine forming methionine (36, 501) (fig. 6). Hammarsten, Reichard and Saluste (73) have shown that in biological systems uracil riboside can be converted to thymine desoxyriboside probably by addition of formate under the control of folic acid (fig. 7). Greenberg (68) has shown that folic acid and formate are essential to complete the ring of 4-amino 5-imidazole carboxamide ribose phosphate, whereby a nucleotide of hypoxanthine, inosinic acid, is formed (fig. 4)

Thus, folic acid activates metabolic processes which lead to the formation of nonessential amino acids; to the transfer of methyl groups to form compounds like methionine; to the methylation of pyrimidine ribosides; and to the closure of the purine ring. It is also probable that folic acid facilitates the incorporation of glycine and formate into the purine ring.

There is additional biologic evidence that folic acid is intimately concerned with the formation of pyrimidines. For instance, Stokes (163) and Snell and Mitchell (154) showed that large amounts of thymine in the presence of purines will replace folic acid as a growth factor for *Streptococcus fecalis* R and *Lactobacillus casei*. Thymine will also overcome the inhibitory effect of methyl folic acid, a folic acid antagonist, on the growth of *L. casei* when purines are available according to Rogers and Shive (130). Furthermore, Prusoff, Tepley and King (126) demonstrated that *L. casei* does

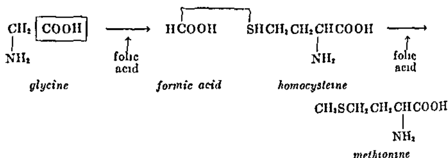


Fig. 6

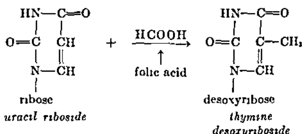


Fig. 7

not form desoxyribose nucleic acid in normal amounts if the folic acid content of the media is reduced This defect can be overcome by the addition of large amounts of thymine but ribose nucleic acid production is increased also. These observations have led to the suggestion that folic acid may act as a coenzyme in the biosynthesis of purines and thymine or their ribotides. Even though there are some divergent observations (121) probably because of differences in the ability of various species of bacteria and animals to utilize free pyrimidines, the weight of evidence favors a

s, their

From-

nicious

anemia will respond to thymine in doses of 8 to 12 grams a day and Vilter,

Horrigan, Mueller, Jarrold, Vilter, Hawkins and Seaman (177) obtained hematopoietic responses with uracil in doses of 30 grams a day. It appears likely that these pyrimidines induce such therapeutic responses by circumventing reactions catalyzed by folic acid.

INTERMEDIARY METABOLISM OF FOLIC ACID AND THE RELATIONSHIP OF ASCORBIC ACID TO FOLIC ACID

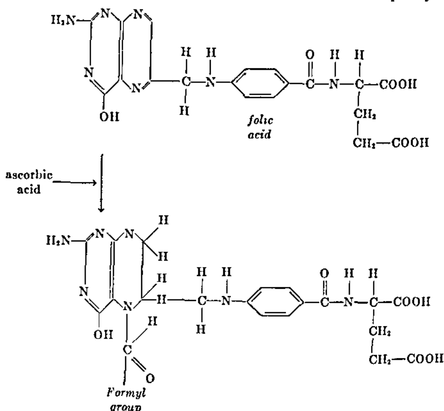
In most foods folic acids occur conjugated with glutamic acid. These are usually the triglutamic acid and the heptaglutamic acid conjugates. Folic acid may be released from such conjugates by conjugases, enzymes which occur in most animal tissues. There are inhibitors for these conjugases in many tissues which block their activity and prevent the release of free folic acid There is evidence that liver extract or vitamin B_{12} protects conjugase from the inhibitor (6).

A form of folic acid called the *citrovorum factor* or *folinic acid* has been found in liver and occurs in rather high concentration in certain liver extracts. It was originally discovered because of its growth promoting effects on an organism *Leuconostoc citrovorum* from which it took its name. Sauberlich and Baumann (137) showed that the citrovorum factor was many times more active in promoting the growth of this organism than was folic acid Later this same factor was recovered by Sauberlich (138) in the urine of animals and human beings following the administration of folic acid Nichol and Welch (116) were able to show that ascorbic acid accelerated the conversion of folic acid to the citrovorum factor or folinic acid in liver slices or in intact laboratory animals and human beings Later Gabuzda, Phillips, Shilling and Davidson (60) showed that folic acid was converted to the citrovorum factor rather poorly in persons with scurvy and that the conversion was greatly increased by the administration of ascorbic acid but only after a lag period of four to five days The citrovorum factor has been available for clinical investigation in the form of leukovorin, crystalline formyl tetrahydro-folic acid (11) (fig. 8). This substance however is probably not identical to the liver citrovorum factor The chemical formula of the latter compound is still unknown. The liver citrovorum factor appears to be even more active metabolically than leukovorin (92). There may be additional folic acid derivatives of even greater biological activity Welch and Heinle (184) have suggested that the final active factor may be a derivative of formyl tetrahydro-folic acid conjugated with ribose phosphate Such a conjugation would make this folic acid coenzyme conform to the configuration of other coenzymes derived from B-complex vitamins—like riboflavin

It is interesting to note that leukovorin is a reduced form of folic acid which contains a formyl (CHO) group Addition or subtraction of this

group may be the means whereby folic acid derivatives act as carriers of single carbon units.

There is an additional relationship between folic acid and ascorbic acid in the oxidation of tyrosine. Scorbutic guinea pigs, monkeys, infants and adult humans excrete parahydroxyphenyl pyruvic acid and parahydroxy-



Synthetic citrovorum factor (leukovorin, folinic acid) (formyl tetrahydrofolic acid)

FIG 8. STRUCTURAL FORMULAE OF FOLIC ACID AND THE CITROVORUM FACTOR AND THE REDUCING EFFECT OF ASCORBIC ACID IN THE FORMATION OF THE CITROVORUM FACTOR

phenyl lactic acid in the urine after ingestion of tyrosine. Ascorbic acid corrects the metabolic defect rapidly (190) Folic acid in large amounts has the same effect in guinea pigs and human infants (64), but not in monkeys or liver slices deficient in ascorbic acid. The relationship to the reactions described above is not clear

METABOLIC FUNCTIONS OF VITAMIN B_{12}

Vitamin B_{12} exists in several different forms, probably with somewhat different quantitative activities as a growth factor for bacteria and animals.

The following forms have been reported: Vitamins B_{12}, B_{12a}, B_{12b} and B_{12c} (26, 153). The generic name, cobalamine (89), has been suggested for the group and seems appropriate, since each variety contains cobalt which gives these compounds a red color. Vitamin B_{12} contains a cyanide radical so it is properly called cyanocobalamine. Vitamins B_{12a} and B_{12b} contain, instead, a hydroxo radical and, therefore, are called hydroxocobalamine. Vitamin B_{12c} is a nitrocobalamine. It is probable that other forms exist with different groups substituted for the cyano group.

The complete chemical structure is not known, though portions of this very large molecule (molecular weight about 1300) have been isolated. They include a dimethyl benzimidazole ring joined to ribose phos-

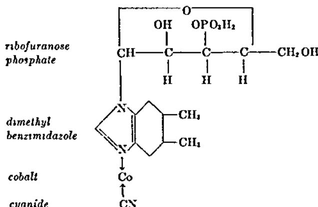


FIG. 9. TENTATIVE PARTIAL STRUCTURE OF VITAMIN B_{12}.

phate in a glucoside linkage; cobalt is probably linked to the benzimidazole radical; two amino propanol groups have been found; and the cobalt is thought to be linked coordinately to the benzimidazole and cyanide, as iron is linked to porphyrin, histidine and oxygen in hemoglobin (fig. 9). A general description of the chemical structure can be found in articles by Emerson and Folkers (53) and by Beaven, Holiday, Johnson, Ellis and Petrow (4).

Vitamin B_{12} is an animal protein factor, that is, it is an essential growth factor for bacteria and animals occurring in animal proteins but not in those from vegetable sources or in yeast Studies in chicks by Gillis and Norris (61) indicated that the animal protein factor spares methyl groups derived from choline. More recently extensive investigation in bacterial nutrition by Davis and Mingioli (36) and in animal nutrition by Dinning, Payne, and Day (45), and Stekol and Weiss (162), reviewed by Jukes and Stokstad (88) have shown that vitamin B_{12} is involved in the synthesis of

methionine, that is, the transfer of a methyl group to homocysteine to form methionine (fig. 10). This methyl group may be derived from choline or betaine, or other sources of labile methyl groups. Once vitamin B_{12} deficiency is induced in animals, and the formation of methionine is suppressed, there is a rather long lag period following administration of vitamin B_{12} before these methylation reactions return to normal This lag period has suggested the possibility that vitamin B_{12} is essential to the formation of an enzyme system necessary for the methylation of homocysteine to methionine, rather than accelerating this reaction itself as folic acid does.

Dubnoff (49) on the basis of work with bacteria and liver slices has suggested that vitamin B_{12} is involved in reactions which activate disulfide (S—S) groups to sulfhydryl (—SH) groups. Since most enzymes have such groups which require activation, such a function would give vitamin B_{12} exceedingly wide biologic importance.

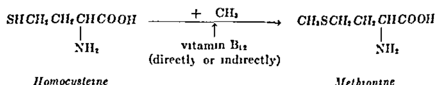


FIG 10

Of even greater interest to this discussion is the relationship of vitamin B_{12} to pyrimidine metabolism Shive, Ravel and Eakin (147) have shown that thymidine, thymine nucleoside, can be substituted for vitamin B_{12} *to promote the growth of Lactobacillus lactis dorner* when purines are available and for the animal protein factor in the nutrition of *Lactobacillus leischmannii* Reisner and West (128) obtained slight reticulocyte responses but no erythrocyte rises in patients with pernicious anemia with 5 to 150 mg. of thymidine, Ungley (169) obtained no response to 50 mg of this material; but Hausmann (76) reported that thymidine induced limited reticulocyte responses and erythrocyte rises in patients with pernicious anemia treated with two to three grams over a ten day period suggesting that in human beings also, thymidine will substitute for vitamin B_{12}

The mechanism by which vitamin B_{12} exerts its effect on the transfer of methyl groups, the activation of (S—S) group and the formation of nucleosides and nucleotides is unknown However, the reactivity of the coordinately linked cyanide group suggests it as a possible means of carbon transfer and the ribose-phosphate moiety raises the possibility that vitamin B_{12} may be acting as a carrier of ribose, or ribose phosphate At any rate, the chemical functions of vitamin B_{12} and folic acid are very closely related.

INTERRELATIONSHIPS OF FOLIC ACID, ASCORBIC ACID AND VITAMIN B_{12}

Experiments summarized above indicate that a most important catalyst derived from folic acid (by way of the citrovorum factor, folinic acid) plays a prominent role in the transfer of single carbon units derived from glycine and serine, particularly methyl and formate groups and that vitamin B_{12} also is involved as an activator of such carbon transfers. By facilitating formate and methyl transfers, folic acid and vitamin B_{12} are probably involved in the formation and closure of purine and pyrimidine rings, particularly that of thymine, the building stones being glycine and other simple amino acids or fragments from them. Vitamin B_{12} seems to play an important part in the formation of nucleosides and nucleotides of these purines and pyrimidines. The formation of the glucoside and ester linkages probably occur before the final step or ring closure. Apparently the most important nucleoside for hematopoiesis derived from these reactions is thymidine (thymine desoxyriboside) and the most important nucleotide is thymidilic acid (thymine desoxyribose phosphate ester or thymine nucleotide). Ascorbic acid acts as a reducing agent in the conversion of folic acid to the citrovorum factor (folinic acid, leukovorin), an intermediate metabolite in the chain of reactions which results in the formation of the active folic acid-like catalyst.

Folic acid, vitamin B_{12} and ascorbic acid play catalytic roles at different levels in these reactions which lead to the formation of nucleic acids. They are not interchangeable. They cannot substitute for each other, but they are dependant upon each other for final completion of the chemical chain reaction which produces the nucleic acids.

STUDIES ON PATIENTS WITH MEGALOBLASTIC ANEMIA WHICH INDICATE THAT THE CHEMICAL REACTIONS PREVIOUSLY DESCRIBED OCCUR DURING HUMAN BLOOD FORMATION

Vitamin B_{12} is metabolically active in pernicious anemia in microgram quantities. Folic acid is effective in milligram doses and thymine and uracil will replace folic acid in doses measured in grams. These discoveries opened the way for investigations concerning the chemical relationship of these substances in human blood formation. The strikingly different amounts of these substances required to induce hematopoietic responses suggests that vitamin B_{12} is a catalyst which is immediately effective at the tissue level, and that folic acid must be converted in the body into a more active product, which also acts catalytically The large doses of thymine and still larger doses of uracil required to induce a hematopoietic effect in patients with pernicious anemia is evidence that these substances function as substrates in a reaction that probably results in the formation of nucleic acids

Thymidine, which replaces vitamin B_{12} in the nutrition of *Lactobacillus leischmanii*, *Lactobacillus lactis*, and some other vitamin B_{12} dependent bacteria, has had limited clinical trials as mentioned previously. If Hausmann's material was pure his data suggest that thymidine is more active hematopoietically than thymine, which in turn is more active than uracil, and that these three substances are substrates in a metabolic chain reaction.

Adenine in daily dose of five grams has been tested by Stone and Spies (164) and we have tested guanine, 2.5 grams, cytidine, 150 mg., uridine, 150 mg., adenosine, 150 mg., and adenylic acid, 50 mg. in patients with pernicious anemia. These materials have been ineffective. These unsuccessful tests coupled with the responses obtained with thymine and its relatives suggest that in pernicious anemia the major defect resides in the formation of pyrimidines of the thymine type. Perhaps the other purines and pyrimidines can be formed easily from this prototype and do not require formation de novo.

Therapeutic tests carried out by us with ribose and desoxyribose nucleic acids in doses of 10 grams daily by mouth have been unproductive Since these substances are split into many smaller molecules during digestion, these tests cannot have much meaning, except to indicate that insufficient amounts of thymine, thymidine, and uracil were liberated to induce a response. They do indicate that the other purine and pyrimidine bases do not have an additive or synergistic effect with thymine and thymidine.

Spectrophotofluorometric, chemical and histochemical studies on erythroblasts from patients with pernicious anemia indicate an abnormality in the cytoplasmic ribose nucleic acid Thorell (167) showed that during the process of maturation of the normal erythrocyte, there is a gradual decrease in the concentration of cytoplasmic ribose nucleic acid as the demands of the dividing cells for new protein diminish. Hemoglobin formation takes place rapidly after ribose nucleic acid has almost disappeared. In erythroblasts from patients with pernicious anemia the concentration of cytoplasmic ribose nucleic acid remains high during hemoglobin formation and does not decrease in a normal manner until after treatment with liver extract

Davidson (31) and Davidson, Leslie and White (32) have studied the nucleic acids of bone marrow cells from patients with pernicious anemia and have found very high values for both ribose and desoxyribose nucleic acids by chemical determination Within several days after treatment with liver extract or folic acid these values fall but the greatest fall occurs in the ribose nucleic acid

Horrigan, Jarrold and Vilter (82) have demonstrated a morphologic change in the ribose nucleic acid of pernicious anemia erythroblasts characterized by clumping and lack of homogeneity in pyronin stained prepara-

tions. Within 48 hours after treatment with vitamin B_{12} or folic acid, the ribose nucleic acid was dispersed throughout the cytoplasm in a normal fashion and disappeared normally as the cells matured.

These observations indicate a defect in the metabolism of cytoplasmic ribose nucleic acid in pernicious anemia erythroblasts. The morphologic evidence indicates an error in breakdown of this substance; the bacteriologic and clinical evidence favors an error in formation. These are compatible since all of these reactions are reversible.

Though the nuclear structure in these cells appears abnormal also (the chromatin is thread-like and nucleoli are present even though hemoglobin is clearly visible in the cytoplasm), neither Reisner and Korson (129) by spectrophotofluorometric methods, nor Horrigan and his associates (82) by cytochemical methods could demonstrate an abnormality in the desoxyribosenucleic acids. Since thymine is a usual component of desoxyribosenucleic acid but not of the ribose type, the clinical therapeutic tests (suggesting an abnormality in thymine-containing desoxyribosenucleic acid) and the chemical determinations (indicating an abnormality in ribose nucleic acid) are not in complete accord. However, both types of investigation demonstrate an abnormality in the metabolism of nucleic acids in erythroblasts of pernicious anemia which is rectified by liver extract, folic acid or vitamin B_{12}.

The Effect of Vitamin B_{12} and Folic Acid in Patients with Pernicious Anemia in Relapse

Both vitamin B_{12} and folic acid induce hematologic remissions in patients with pernicious anemia in relapse, a disease characterized by megaloblastic anemia, permanent achylia gastrica, posterolateral column degeneration and peripheral neuritis, indirect reacting bilirubinemia, glossitis and other gastro-enteric tract abnormalities There are minor differences in these responses. Vitamin B_{12} in microgram doses given parenterally usually induces rapid improvement in sense of well being, rapid rise and fall in numbers of reticulocytes with the sharp peak occurring on the eighth or ninth day, rapid rise in erythrocytes and hemoglobin, rapid disappearance of bilirubin from the plasma, relief of glossitis accompanied by papillary regeneration and slow improvement in the function of peripheral and central nervous systems

The response to folic acid is the same whether it is given orally or parenterally in milligram doses. Most of the signs of improvement occur more slowly Often there is rapid deterioration in central or peripheral nerve function though occasionally one sees a patient who shows temporary subjective and objective improvement The reticulocytosis following folic acid administration usually is slower to appear, slower to reach a maximum

and slower to fall. Commonly one observes a secondary increase in reticulocytes after the first response with no change in the dose of folic acid Occasionally one finds a patient with pernicious anemia who responds poorly to 10, 20 and even 30 mg. of folic acid a day but responds well to a few micrograms of vitamin B_{12} These therapeutic observations suggest that the mode of action of these two agents, though very similar, is not identical.

If one continues to treat one group of patients with vitamin B_{12} and another with folic acid, additional differences become apparent as reported by Vilter and associates (177). The patients treated with 10 micrograms of vitamin B_{12} every two weeks or 20 micrograms monthly remain well hematologically and neurologically though Beard (3) noted persistence of macrocytosis in such patients on diets poor in animal protein. More than ⅔ of those patients treated with folic acid in doses of 10 to 15 mg. daily or 30 mg. three times a week develop neurologic relapse in 12 to 18 months (174, 78). A few patients whose neurologic symptoms remain at a minimum, develop hematologic relapse within two to three years (177). Similar hematologic relapses have been noted by Schwartz, Kaplan and Armstrong (143) and Hansen-Pruss (74). Such a relapse is characterized by macrocytic anemia, leukopenia, thrombopenia, and a typical megaloblastic bone marrow. Response may be obtained rapidly with refined liver extract, vitamin B_{12}, thymine or by increasing the dose of folic acid to 50 mg. daily. Patients treated with thymine or folic acid will not maintain this remission for long Usually in two to eleven months, another hematologic relapse has occurred which will usually respond to increases in the folic acid dosage to 100 mg. daily. Usually in the second relapse and almost always by the third, the bone marrow has become hypocellular and inactive with few clear evidences of megaloblastic maturation arrest though the peripheral blood remains macrocytic. Invariably signs of degeneration in the peripheral and central nervous system have been present Hematologic response to refined liver extract or vitamin B_{12} is very slow or is absent, though some response to thymine still can be obtained. Vitamin B_{12} combined with a high protein diet results in return of the bone marrow to a normally active appearance and to regeneration of blood, but only after three to six months of continuous treatment In striking contrast to the slow improvement in the peripheral blood and bone marrow, vitamin B_{12} induces very rapid relief of neurologic symptoms and signs. Most of the disabling manifestations disappear in two to three weeks (177)

These observations indicate clearly that the chemical reactions induced in patients with pernicious anemia by folic acid and by vitamin B_{12}, though related, are not identical It is likely that folic acid induces hematologic remissions in persons with pernicious anemia by "mass action" effect, for the same dose that induced the remission will not maintain it indefinitely. When relapse occurs, thymine or an increase in the dose of folic acid will

induce another temporary remission. The concept of "mass action" in turn suggests that folic acid is a substrate in the formation of another biologically active catalyst which accelerates the synthesis of pyrimidines of the thymine type. Under such circumstances each increase in the dose of folic acid tends to deplete the body of enzymes necessary for such a conversion. The hypoplastic marrow which occurs in patients with pernicious anemia treated with increasingly larger doses of folic acid is reminiscent of aminopterin induced bone marrow hypoplasia, suggesting that depletion of the body's reserves of these enzymes by continuous treatment with folic acid may induce as severe a deficiency in the folic acid coenzyme as can be obtained by administration of aminopterin. If both folic acid and vitamin B_{12} accelerate closely related dependent chemical reactions, the scant supply of vitamin B_{12} available to persons with pernicious anemia will be depleted also. Such depletion of vitamin B_{12} may be the reason why every patient developed neurologic relapse as the dose of folic acid was increased to combat hematologic relapse. There is no good explanation, however, for the almost equal initial hematologic response to both vitamin B_{12} and folic acid in pernicious anemia (a conditioned vitamin B_{12} deficiency), unless, as has been suggested many times, the deficiency of vitamin B_{12} induces a deficiency in folic acid as a secondary manifestation. These relationships are indicated in diagrammatic form in figure 11.

Direct Effect of Vitamin B_{12} and Folic Acid on the Megaloblasts of Pernicious Anemia

Horrigan, Jarrold and Vilter (82) demonstrated that one microgram of vitamin B_{12} injected into the iliac crest marrow cavity of patients with pernicious anemia, induced maturation of the megaloblastic bone marrow to a normoblastic type at the site of injection but not in the opposite iliac crest. Folic acid, at a one or two mg level or the citrovorum factor in equivalent amounts did not have this effect. Only when folic acid was given in sufficient quantity to induce a general hematologic response was the morphology of the marrow changed to a normoblastic type. These observations were interpreted to mean that vitamin B_{12} could be utilized by the bone marrow cells without alteration, that enzyme systems were present in the megaloblast which lacked only vitamin B_{12} for potentiation. Folic acid on the other hand had to be converted into a biologically active compound elsewhere in the body—it could not be utilized in the form of folic acid by the megaloblast, perhaps because it diffused out of the marrow too quickly. These observations are consistent with the hypothetical reaction illustrated in figure 11. Vitamin B_{12} is apparently active as such, folic acid must be converted to an unknown related compound before it acquires biologic activity.

However, Lajtha (97) and Callender and Lajtha (15) have reported that

Coenzymes

Folic acid conjugates → Folic acid → Folinic acid (a citrovorum factor) (leukovorin) → Unknown active factor(s)

Conjugases (B_{12} protects?) ↑ (Folic acid conjugates → Folic acid)

Ascorbic acid ↑ (Folic acid → Folinic acid)

↗ ?

Substrates

Methyl (CH_3), formate (HCOOH) and amino (NH_2) groups from glycine or serine and other simple compounds → Incomplete pyrimidine and purine rings + Pentose or pentose phosphate → Precursors of pyrimidine and purine nucleosides and nucleotides + Methyl or formate → Pyrimidine and purine nucleosides and nucleotides → Nucleic acids

Uracil or thymine will substitute ↑ (Incomplete pyrimidine and purine rings)

Vitamin B_{12} ↓ (→ Precursors of pyrimidine and purine nucleosides and nucleotides)

Thymine will substitute ↑ (Precursors of pyrimidine and purine nucleosides and nucleotides)

closure of the ring

These are probably reversible reactions and account for both the synthesis and degradation of nucleic acid

Later in this paper this reaction will be related to human blood formation and the origin of the macrocytic anemias.

FIG. 11. SCHEMATIC REPRESENTATION OF REACTIONS IN NUCLEIC ACID FORMATION CATALYZED BY FOLIC ACID, VITAMIN B_{12} AND ASCORBIC ACID.

pernicious anemia megaloblasts cultured in pernicious anemia serum by a modified Osgood technique in vaccine vials mature to normoblasts when folic acid or citrovorum factor is added. In their experiments vitamin B_{12} was inactive until combined with normal gastric juice. These two experiments seem to be irreconcilable at first glance. However, both vitamin B_{12} and folic acid may be found to be active in the test tube when optimal conditions are available and the failure of folic acid to induce local maturation of the iliac crest may turn out to be due to the rapid diffusion of this crystalloid material out of the marrow before it can be converted to a metabolically active substance as in figure 11. Vitamin B_{12}, a very large molecule and one that conjugates readily with many proteins, (131) may be retained in the marrow and exert its catalytic effect without further alteration.

Studies in Patients with Macrocytic Megaloblastic Anemia That Responds Poorly to Vitamin B_{12}

There are patients with macrocytic anemia, megaloblastic bone marrow, normal serum bilirubin, absence of spinal cord disease and free hydrochloric acid in the gastric juice who respond poorly, if at all, to refined liver extract or vitamin B_{12} but respond well to folic acid When such anemia occurs in an infant it is called megaloblastic anemia of infancy; when in a pregnant woman, pernicious anemia of pregnancy, when in an adult male or non-pregnant woman it is called liver extract refractory megaloblastic anemia or *achrestic* anemia. Patients who fall in these categories must have a metabolic abnormality involving folic acid for they have a rapid hematologic response to folic acid, but are usually refractory to vitamin B_{12} (34, 35, 186, 179, 194, 150) Hematologic study of these patients has thrown some light upon the intricacies of folic acid metabolism

Megaloblastic Anemia of Infancy. Infants with megaloblastic anemia usually have been fed a processed milk diet unsupplemented with ascorbic acid About twenty-five per cent of them have clinical scurvy indicative of the severe degree of ascorbic acid deficiency induced by this diet. Analyses show that the milk diet is deficient also in folic acid On the basis of such information, May, Nelson, Lowe and Salmon (102) succeeded in producing a similar clinical syndrome in monkeys by feeding a milk diet low in vitamin C and folic acid These monkeys developed macrocytic anemia, megaloblastic maturation arrest in the bone marrow, and clinical scurvy. The megaloblastic bone marrow became normoblastic and the anemia improved when folic acid was given but scurvy was unaffected until ascorbic acid was administered. Ascorbic acid alone in the absence of folic acid relieved the scurvy but the anemia responded slowly or not at all Vitamin B_{12} alone had no therapeutic effect but when administered in conjunction with ascorbic acid, it induced rapid erythrocyte regeneration. Monkeys

fed this diet with supplements of folic acid developed scurvy, a normocytic normochromic anemia, and a normoblastic bone marrow. The diet supplemented with ascorbic acid was apparently not sufficiently deficient in folic acid to induce anemia or megaloblastosis. The milk diet induced megaloblastic anemia in monkeys because it was deficient both in folic and ascorbic acid and there is good evidence that a similar combined deficiency induces megaloblastic anemia in infants. Since manufacturers of processed powdered milks have been adding ascorbic acid to their formulae, megaloblastic anemia of infancy has almost disappeared.

The fact that a combined deficiency of folic acid and ascorbic acid induces a more profound deficiency in folic acid than can be obtained by folic acid deficiency alone is compatible with the relationship of ascorbic acid to the conversion of folic acid to the citrovorum factor (folinic acid). Nichol and Welch (116) showed that ascorbic acid had a profound effect on this conversion *in vitro* and *in vivo*, and Gabuzda, Phillips, Shilling and Davidson (60) demonstrated that scorbutic patients were unable to excrete much citrovorum factor in the urine in response to a test dose of folic acid until after the scurvy was relieved by ascorbic acid. Because of these relationships citrovorum factor was tried in monkey megaloblastic anemia (101) and found to be many times more effective than an equivalent dose of folic acid

These considerations support the hypothesis that ascorbic acid is essential for one step in the conversion of folic acid to its metabolically active form. Leukovorin (a citrovorum factor, tetra hydroformyl-folic acid) is not this final active form because Meyer, Brahin and Sawitsky (105), Horrigan, Jarrold and Vilter (82), and Ellison, Wolfe, Lichtman, Ginsberg and Watson (51) showed that it was no more effective in pernicious anemia than folic acid and it was inactive in those patients with pernicious anemia who had become refractory to folic acid (82).

These data also serve to explain the occasional adult patient who has macrocytic megaloblastic anemia associated with scurvy There are several such cases in the literature reported by Vilter, Woolford and Spies (175) and by Jennings and Glazebrook (86) In these cases the anemia and megaloblastic bone marrow were improved rapidly by ascorbic acid. Prior administration of liver extract had failed to induce a remission in the anemia (86)

These data also help explain the occasional patient with nutritional macrocytic anemia (dietary deficiency of vitamin B_{12} and folic acid) who responds to massive doses of vitamin C (176) In all these cases, apparently, vitamin C increases the efficiency of small amounts of folic acid already available to the patient in his diet

Pernicious Anemia of Pregnancy A patient with pernicious anemia

of pregnancy reported by Vilter and his associates (177), after failing to respond to vitamin B_{12}, refined liver extract and uracil, responded to the administration of methionine and choline (sources of labile methyl groups) and thymine. The authors felt that this patient had a defect in the conversion of folic acid to its metabolically active form. They also proposed that one of the functions of folic acid in this patient was acceleration of methylation reactions, one of which was the methylation of uracil-like compounds to substances related to 5-methyl uracil (thymine), a function of folic acid to which reference has already been made. These observations recall earlier reports by Davis and Brown (37) that choline promotes hematologic responses in patients with pernicious anemia and related macrocytic anemias. We have not been able to demonstrate, however, hematologic response in patients with pernicious anemia treated with choline, methionine or betaine (a lactone formed by the action of choline oxidase upon choline).

Since monkey megaloblastic anemia responded to ascorbic acid and vitamin B_{12}, but not to B_{12} alone, Holly (81) treated three patients with pernicious anemia of pregnancy with these two vitamins. He demonstrated that neither substance alone had beneficial effect on the anemia but when they were given in combination, recovery was rapid. Such observations are difficult to explain with our present knowledge. One might postulate a combined deficiency of folic acid, ascorbic acid and vitamin B_{12} in these patients, and presume that sufficient folic acid was activated by ascorbic acid to induce maturation of the erythroblasts when the deficiency of B_{12} was rectified This would presuppose that neither vitamin B_{12} nor ascorbic acid were stored in the body which of course is incorrect On the other hand, it is possible that vitamin B_{12} as well as ascorbic acid activate folic acid, the former by the addition of ribose or a single carbon unit, the latter by reduction. Whether either or these hypotheses is correct remains to be seen.

Liver Extract Refractory Megaloblastic Anemia (Achrestic Anemia This type of anemia is rather rare in the United States For unknown reasons the disease occurs in Northern European countries much more frequently. A patient reported by Mueller, Hawkins and Vilter (113) failed to respond to refined liver extract, vitamin B_{12}, uracil, choline and methionine and ascorbic acid, but did respond dramatically to folic acid or thymine. This patient excreted a *normal amount* of folic acid in the urine before therapy and responded to a load test as though he had normal folic acid stores in his tissues. He probably had an abnormality in the metabolism of folic acid somewhat similar to that of patients with pernicious anemia of pregnancy. This defect impaired the ability of the body to form thymine-like substances

Very rarely one finds a patient with megaloblastic anemia who fails to respond to all the types and combinations of therapy previously mentioned The authors have had the opportunity to study such a patient over a period of eighteen months. He failed to respond to vitamin B_{12} given orally or parenterally, to refined or crude liver extract given parenterally, to crude oral liver extract (Valentine's) and to folic acid, xanthopterin, thymine, uracil, methionine and choline. Eventually he developed pneumonia and died. At necropsy his bone marrow was almost completely acellular and fatty, though up to his terminal illness it had been megaloblastic and at the beginning of the study, hyperblastic. It must be presumed that such a patient has some complicated defect in the metabolism of nucleic acids than can not be explained with our limited knowledge at present.

Sprue and Nutritional Macrocytic Anemia

These two disorders are usually characterized by megaloblastic macrocytic anemia, free hydrochloric acid in the gastric juice after histamine, absence of spinal cord disease and normal serum bilirubin. Both respond to folic acid (28, 157) or vitamin B_{12} (158, 159) given orally or parenterally. Both diseases occur in chronically malnourished persons. They differ only in the type of diarrhea Sprue is characterized by steatorrhea while patients with nutritional macrocytic anemia usually have a watery diarrhea In monkeys, folic acid deficiency is very similar to sprue in its clinical manifestations (28). Nutritional macrocytic anemia is probably similar to extrinsic factor deficiency in pigs. It is probable that they are combined dietary deficiencies of folic acid and Vitamin B_{12}. In sprue the folic acid deficiency predominates; in nutritional macrocytic anemia, the principal dietary deficiency is vitamin B_{12}.

Other Types of Megaloblastic Anemia

Megaloblastic anemia which has been reported from the tropics (tropical macrocytic anemia) occurs most commonly in pregnant women and is probably related to pernicious anemia of pregnancy It is said to respond well to folic acid but poorly to vitamin B_{12} (187) Macrocytic anemias due to bowel resection, short-circuiting operations and gastro-colic fistulae will be referred to later. They respond well to folic acid (112). The fish tape worm anemia, probably due to inactivation of intrinsic factor by the worm responds in a fashion identical to pernicious anemia (178).

EXPERIMENTAL DEFICIENCIES IN ANIMALS WHICH SUPPORT THESE RELATIONSHIPS

Cartwright, Tatting, Ashenbrucker and Wintrobe (18) and Heinle, Welch, George, Epstein and Prichard (79) have induced nutritional macro-

cytic anemia in pigs by a vitamin free casein and synthetic nutrient diet with or without a crude folic acid antagonist. These pigs were deficient principally in folic acid though extrinsic factor (vitamin B_{12}) was diminished. The bone marrow of these animals contained cells very similar to pernicious anemia megaloblasts The hematopoietic response to refined liver extract (vitamin B_{12}) was poor. When a response did occur it was short lived and a relapse soon followed Optimal improvement occurred only when folic acid was given. This effect is similar to that observed by Vilter and his associates in patients with pernicious anemia who relapsed on folic acid (177). The primary deficiency in the patients with pernicious anemia is vitamin B_{12}, and improvement with folic acid is only temporary. In the pigs folic acid was deficient, and the response to vitamin B_{12} was unsatisfactory. These observations indicate that both folic and vitamin B_{12} are essential for hematopoiesis.

Heinle, Welch and Shorr (80) and later Cartwright and his associates (19) have attempted to induce vitamin B_{12} deficiency in pigs on a synthetic diet containing soybean protein. None of these animals developed megaloblastic bone marrow, or macrocytosis. Heinle and his associates demonstrated reticulocytosis following administration of vitamin B_{12}. Cartwright's animals responded principally with weight gain when vitamin B_{12} was administered. This is a rather discouraging result of an attempt to induce a state in pigs comparable to pernicious anemia. Either an insufficient degree of deficiency was induced in these animals or there are defects other than a conditioned deficiency of vitamin B_{12} in pernicious anemia.

Dietrich, Nichol, Monson and Elvehjem (40) and Dietrich, Monson and Elvehjem (41) have tried to define some of the relationships of vitamin B_{12}, folic and citrovorum factor (folinic acid) in chicks. They have shown that the livers of folic acid deficient chicks contained increased amounts of folic acid when the chicks were treated with small doses of vitamin B_{12} parenterally Larger amounts, given parenterally, had an inhibitory effect, that is, there was less folic acid but the same amount of citrovorum factor. Orally administered vitamin B_{12} increased the level of both folic acid and citrovorum factor in these livers The same effect can be obtained by giving vitamin B_{12} parenterally and adding ascorbic acid to the livers *in vitro*

Conversely, chicks made deficient in vitamin B_{12} have more of this substance in their livers after treatment with folic acid. These effects may indicate a relationship of one of these vitamins to the formation of the other, or they may be the result of the action of vitamin B_{12} or folic acid on the ability of the intestinal organisms of the chicks to synthesize the deficient vitamin which then is stored in the liver.

Such considerations raise the question of the possible effects of gastro-

intestinal tract organisms on the availability and metabolism of folic acid and vitamin B_{12} in animals and human beings. There are a number of hypothetical possibilities. These organisms may absorb so much of one of these vitamins that a deficiency occurs. On the other hand they may, under various stimuli, manufacture more of these vitamins which then become available to the host. An even more complicated relationship is possible. Because of the preexisting abnormality in the stomach or bowel, an abnormal intestinal flora may become established with organisms which absorb more of one of these vitamins or produce inhibitory substances or metabolic antagonists for these vitamins. Thompson (166) and Lajtha (97) have reported such inhibitors in the serum of persons with pernicious anemia. Lichtman, Ginsberg and Watson (99) have reported that aureomycin has limited reticulocytogenic effects in pernicious anemia, presumably because this antibiotic destroys objectionable gastro-intestinal tract organisms or allows more favorable ones to multiply. The megaloblastic anemia in animals and man induced by intestinal short-circuiting operations, blind intestinal loops and gastro-colic fistulae is probably due to abnormalities in the organisms of the gastro-intestinal tract and their effects on vitamin B_{12} and folic acid metabolism.

THE EFFECT OF FOLIC ACID ANTAGONISTS ON ANIMAL AND HUMAN BONE MARROW CELLS

Aminopterin and amethopterin, two very potent antagonists of folic acid which have been used in the treatment of acute leukemias, induce complete aplasia of the marrow of animals or human beings when given in doses slightly in excess of the therapeutic levels This occurs more rapidly if the animal or human being is deficient in folic acid or related catalysts. These substances also interfere with the growth of the cells of acute leukemia in mice or in children and may induce a temporary remission in the disease. Occasionally megaloblasts have been observed in the marrows of leukemic persons treated with aminopterin. However, Burchenal, Johnston and Waring (13) and Burchenal, Webber and Johnston (14) have shown that leukemic cells of mice can develop resistance to these antagonists by repeated passage through mice treated with one of these drugs Presumably the same thing happens in children because eventually these drugs lose their effectiveness and the leukemic process spreads like wildfire in spite of them. The mechanism whereby resistance to aminopterin develops is not defined clearly but it has its counterpart in the ability of bacteria to develop resistance to antibiotic drugs presumably through mutation and survival of resistant mutants Resistant cells either develop alternate metabolic pathways which avoid the reaction blocked by the

antagonist or they develop the ability to metabolize the antagonist and convert it to their own purposes.

Aminopterin has been shown to exert its effect by competition not with folic acid but with the metabolic process which produces citrovorum factor (117). Schoenbach, Greenspan and Colsky (142) showed that a few milligrams of leukovorin, a crystalline citrovorum factor, will reverse the effect of one mg. of aminopterin whereas at least 200 to 600 mg. of folic acid are required to have the same effect. Presumably these antagonists induce a deficiency in the biologically active derivatives of folic acid and in this way interfere with metabolism of nucleic acids, and the transport of single carbon units

RECAPITULATION

Results of therapeutic tests and other experimental procedures on patients with megaloblastic anemias indicate that the chemical effects of vitamin B_{12}, folic acid and ascorbic acid on the growth and maturation of human erythroid cells are much the same as those which occur during bacterial growth and *in vitro* metabolism of liver slices. Abnormalities in these reactions result in defective blood formation and anemia. These are reversible reactions leading to the formation of the nucleic acids. In essence, they constitute a chain reaction, dependent at different stages upon both folic acid and vitamin B_{12}

When vitamin B_{12} is absorbed from the gastro-intestinal tract through the action of the stomach enzyme, the intrinsic factor, it is effective immediately and without change in chemical constitution It catalyzes the transfer of single carbon units and appears to facilitate the formation of nucleosides and nucleotides, perhaps by catalyzing the transfer of ribose phosphate to precursors of the purine and pyrimidine rings. It has something to do with the activation of enzymes containing (S—S) groups

On the other hand folic acid must be converted to a metabolically more active substance or substances probably through the citrovorum factor as an intermediate stage. Ascorbic acid, acting as a reducing agent, speeds up this reaction and aminopterin competitively inhibits it. Vitamin B_{12} also may have a direct effect upon it Folic acid in its active coenzyme form is essential for the formation of the pyrimidine and purine rings, probably through the part it plays in the transfer of single carbon units There are probably additional catalysts than those already mentioned which are essential to these chemical reactions, and these substances as well as vitamin B_{12} may be depleted when a patient with pernicious anemia, a conditioned vitamin B_{12} deficiency, is treated with folic acid.

A deficiency of any of these substances may induce a megaloblastic

anemia. Absence of any of them may cause marrow aplasia and death. The megaloblast of pernicious anemia appears to be a normal red cell precursor (an erythroblast) with an abnormality in the nucleic acids because of a defect at one of the steps in the formation or degradation of these substances.

These studies also suggest that the several varieties of megaloblastic

TABLE 1

The Clinical Varieties of Megaloblastic Anemias Together with the Probable Deficiency Involved in the Etiology of Each and the Treatment Most Likely to Be Effective

DISEASE	DEFICIENCY	TREATMENT
Pernicious anemia	Vitamin B_{12} conditioned by a deficiency of intrinsic factor	Vitamin B_{12} parenterally
Nutritional macrocytic anemia	Vitamin B_{12} and folic acid (dietary), the former deficiency predominating	Vitamin B_{12} or folic acid
Sprue	Vitamin B_{12} and folic acid (dietary), the latter deficiency predominating	Folic acid
Pernicious anemia of pregnancy	Folic acid coenzyme due to metabolic abnormality	Folic acid
Megaloblastic anemia of infancy	Folic acid and ascorbic acid (dietary)	Folic acid and ascorbic acid
Refractory megaloblastic anemia	Folic acid coenzyme due to metabolic abnormality	Folic acid
Megaloblastic anemia of scurvy	Ascorbic and folic acids (dietary)	Ascorbic acid and folic acid
Megaloblastic anemia due to intestinal blind pouches, etc	Folic acid coenzymes due to abnormality induced by G I tract organisms	Folic acid, aureomycin and surgery
Fish tapeworm anemia	Vitamin B_{12} due to inactivation of intrinsic factor by the worm	Vitamin B_{12} and vermifuge

anemia are etiologically related to a single or combined deficiency state as indicated in table 1.

Not only do deficiencies of folic acid and vitamin B_{12} adversely affect blood formation, such deficiencies also induce abnormalities in the tongue, gastroenteric tract and central nervous system Glossitis, stomatitis and gastritis occur in either deficiency. Steatorrhea and poor absorption of all nutrients from the gastro-intestinal tract are peculiar to folic acid deficiency but may be less apparent results of vitamin B_{12} deficiency also Though peripheral neuritis is common to several deficiency states, posterolateral column disease, including peripheral neuritis is almost specific for vitamin B_{12} deficiency and, with very rare exceptions, occurs only in per-

nicious anemia. It is probable that all organs and tissues of the body are adversely affected by these deficiencies but the most striking evidence of lack is observed in the bone marrow, gastro-intestinal tract and central and peripheral nervous systems. In a similar fashion, other metabolic abnormalities probably affect the formation of phospholipids, proteins and the respiratory enzymes of the erythroid cells The result may be anemia of unrecognized etiology Studies in the metabolism of these erythroblast constituents may uncover chemical reactions even more complex than those reported in this review and will almost certainly lead to the development of new therapeutic agents.

The Chemistry of Hemoglobin Formation

Hemoglobin is by far the most important constituent of the mature red blood cell, comprising about 95 per cent of the cell dry weight The factors which go together to form this complex protein of molecular weight 68,000 are numerous but we will discuss only those of major importance, i.e., protoporphyrin, globin and iron. The colorless portion of the molecule is made up of globin which has attached to it four prosthetic groups of colored heme molecules. Heme itself which is present in many other enzymes such as catalase or peroxidase, is composed of protoporphyrin 9, type III with an iron atom occupying a center position (fig 12) The enzymes or coenzymes necessary for the combination of these substances to form hemoglobin are little understood

PROTOPORPHYRIN 9 METABOLISM

This molecule, originally named by Hans Fischer, is composed of four pyrrole rings joined by —CH methene bridges This naturally occurring protoporphyrin is distinguished by the presence of four methyl groups ($—CH_3$), two vinyl groups ($—CH = CH_2$) and two propionic acid rests ($—CH_2\,CH_2\,COOH$) replacing the eight hydrogen atoms of the porphyrin molecule (fig. 12) Granick and Gilder (65), have shown through studies utilizing the organism *Hemophilus influenza* that the vinyl groups are apparently essential for incorporating iron into the porphyrin ring and that the propionic acid groups help orient the heme in relation to globin as well as help attach the heme to globin In addition, evidence is offered that the porphyrin ring is completely formed before iron enters and that it is not put together part by part about the iron atom. The two negatively charged carboxyl groups of the propionic acid rests are thought to attach to two positively charged groups of globin, possibly the guanidino groups of arginine. Two other naturally occurring porphyrins which are of interest clinically, coproporphyrin and uroporphyrin, differ from protoporphyrin merely in the number and type of side chains attached to the porphyrin ring

Until recently it had been assumed that the porphyrin molecule was derived from pre-formed pyrrole rings existing in compounds such as tryptophane, proline or oxyproline. However, through the isotope studies of Shemin, London and Rittenberg (145) it is apparent that the N atoms of the pyrrole rings are obtained from the N of glycine which also provides four alpha carbons to the pyrrole rings as well as four alpha carbons for the methene bridges. Furthermore, another simple compound, acetate, provides a methyl carbon which is converted to the methyl and beta carbons of heme. The two carboxyl groups in the propionic acid rest also come from the carboxyl groups of acetate. Thus, it is obvious that the protoporphyrin of hemoglobin is built from comparatively simple substances.

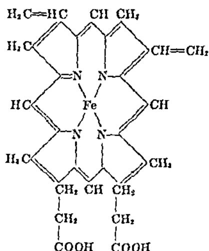


Fig. 12 Chemical Structure of Heme

This explains the ease with which the body manufactures hemoglobin and the rarity with which we see clinically any deficiency in hemoglobin produced by a defect in protoporphyrin synthesis.

Plaut, Betheil and Lardy (122) have suggested that folic acid may play a definite role in the synthesis of heme, possibly through its capacity to transfer single carbon units. The authors, utilizing C^{14} labeled formate, were able to recover the labeled carbon in heme in those animals receiving folic acid, but not in folic acid-deficient animals. However, the exact position of C^{14} in heme was unknown, and whence it came was conjectural. It may well have arrived by way of serine through pyruvate to acetate. On the other hand, it may have been a direct incorporation of C^{14} into heme and if so folic acid has yet another specific function in hematopoiesis.

From the above experiments a working hypothesis has been derived for the biosynthesis of protoporphyrin. This biosynthesis presumably takes

place within the erythroblast and is completed by the time the mature erythrocyte is formed. Since most, if not all, of the carbon atoms of the protoporphyrin molecule are derived from glycine and acetate and since the latter is converted into pyruvate, it is possible that these substances are converted into α-ketoglutaric acid via the tricarboxylic acid cycle Two

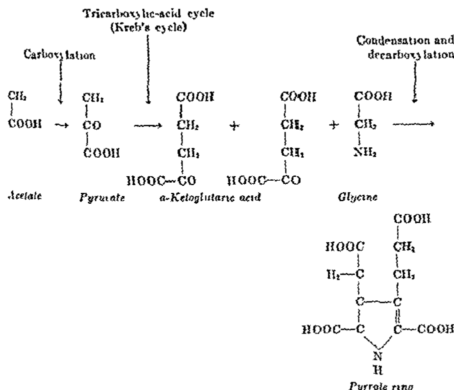


FIG 13 SCHEMATIC REPRESENTATION OF A SERIES OF REACTIONS WHEREBY A PYRROLE RING MAY BE FORMED FROM ACETATE AND GLYCINE

See discussion

molecules of α-ketoglutaric acid condense with one molecule of glycine to form a pyrrole after decarboxylation (fig 13) Four of these pyrroles are then joined by methene bridges to form uroporphyrin, type III By further decarboxylation and dehydrogenation coproporphyrin, type III is formed, and from it arises protoporphyrin For a more detailed account of this hypothesis the reader is referred to the work of Neuberger, Muir and Gray (115) or Lemberg and Legge (98). Shemin and Wittenberg (146) have recently reported evidence obtained by utilizing isotopic C^{14} which indicates that the precursor of the pyrrole ring may not be α-ketoglutaric acid,

Until recently it had been assumed that the porphyrin molecule was derived from pre-formed pyrrole rings existing in compounds such as tryptophane, *proline or oxyproline. However, through the isotope studies of* Shemin, London and Rittenberg (145) it is apparent that the N atoms of the pyrrole rings are obtained from the N of glycine which also provides four alpha carbons to the pyrrole rings as well as four alpha carbons for the methene bridges Furthermore, another simple compound, acetate, provides a methyl carbon which is converted to the methyl and beta carbons of heme. The two carboxyl groups in the propionic acid rest also come from the carboxyl groups of acetate. Thus, it is obvious that the protoporphyrin of hemoglobin is built from comparatively simple substances.

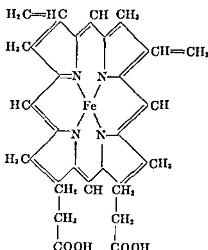


FIG. 12 CHEMICAL STRUCTURE OF HEME

This explains the ease with which the body manufactures hemoglobin and the rarity with which we see clinically any deficiency in hemoglobin produced by a defect in protoporphyrin synthesis.

Plaut, Betheil and Lardy (122) have suggested that folic acid may play a definite role in the synthesis of heme, possibly through its capacity to transfer single carbon units. The authors, utilizing C^{14} labeled formate, were able to recover the labeled carbon in heme in those animals receiving folic acid, but not in folic acid-deficient animals. However, the exact position of C^{14} in heme was unknown, and whence it came was conjectural. It may well have arrived by way of serine through pyruvate to acetate On the other hand, it may have been a direct incorporation of C^{14} into heme and if so folic acid has yet another specific function in hematopoiesis.

From the above experiments a working hypothesis has been derived for the biosynthesis of protoporphyrin. This biosynthesis presumably takes

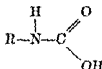
feces. Recently Schmid and Watson (141) have shed more light on the relationship of congenital porphyria to blood formation. They studied thirteen cases, two of the congenital photosensitive type, six of the acute intermittent type and five of the mixed variety. The normoblasts of the two cases of congenital porphyria exhibited intense red fluorescence under the microfluorospectroscope, particularly in the nuclei, which indicated that porphyrin synthesis may take place in part at this site. Uroporphyrin was crystallized from the bone marrow in these cases Furthermore, hemolytic anemia was a rather constant feature of this disease. However, in the remaining eleven cases, porphyrins of the bone marrow were normal. It was felt by these workers that congenital porphyria was indeed directly related to hemoglobin formation and might well be classified as a dyscrasia of the blood forming organ (porphyria erythropoietica). The lack of anemia in most cases of porphyria is not surprising if one considers that in this disease there is an overproduction of porphyrins in addition to an aberration in their formation Thus adequate protoporphyrin is available for hemoglobin formation.

GLOBIN METABOLISM

This protein has a molecular weight of 66,000 and is composed of a mixture of amino acids. The exact make-up is unknown but among the more important amino acids are histidine, arginine, tryptophane, lysine and proline

Globin contains more histidine than most proteins Provision of an imidazole nitrogen which attaches to iron appears to be one of the most important functions of this amino acid. This will be discussed in detail later. The histidine residues also serve as excellent buffers as has been shown by Wyman (192).

Arginine supplies the guanidino groups to which the carboxyl groups of the propionic acid rests of protoporphyrin attach (125) It has been suggested that about 20 to 30 per cent of the total CO_2 is transported by the erythrocyte in the form of carbamino compounds such as



For this to occur it has been postulated that the amino group of valine would have to shift by some two pH units to the acid side (132).

The formation of globin is assumed to occur in the erythroblast and is contributed to by the same donors as all other complex proteins in the body Nothing more of its metabolism or manufacture is known.

but possibly an oxidative decarboxylation product of the latter or a succinyl-coenzyme complex derived from succinate via the tricarboxylic acid cycle.

Most of the protoporphyrin formed is immediately combined with iron and globin to form hemoglobin, but a significant and measurable amount remains in a free state within the erythrocyte Normally this is generally less than 40 micrograms per 100 cc of erythrocytes. Deviations from normal are usually upward and are generally what one would expect. Thus in iron deficiency where iron is not available for the formation of heme, the free erythrocyte protoporphyrin (E.P.) is elevated Also it is increased to high levels in lead intoxication and the anemia of infection both of which are related to poor utilization of iron. E.P may be moderately elevated in hemolytic anemia, leukemia, Hodgkin's disease and multiple myeloma. The fact that the E.P. increases in a sterile preparation of erythrocytes incubated *in vitro* suggests that protoporphyrin synthesis can occur in the mature red cell (181)

The study of free erythrocyte protoporphyrin has led to a hint about the enzymes necessary for the biosynthesis of protoporphyrin Pyridoxine deficiency in some animals can induce a hypochromic anemia *In this* instance the E.P. is low, yet the serum iron is very high, suggesting some abnormality in the production of protoporphyrin (189). Furthermore, pyridoxal phosphate, the active coenzyme of pyridoxine, is known to function as a co-decarboxylase Thus, pyridoxine may occupy an important position in hemoglobin formation. Studies of pyridoxine deficiency in human beings induced with desoxypyridoxine, a metabolic antagonist of pyridoxine, failed to reveal anemia attributable to the deficit of vitamin B_6 (114) but these were acute experiments not sufficiently long to eliminate a relationship *between pyridoxine and human blood formation*

From the foregoing it is not surprising that free coproporphyrin has been found in minute amounts (two micrograms per 100 cc. of erythrocytes) in mature human red blood cells (182). Uroporphyrin may also exist but as yet has not been measured. The free coproporphyrin may be correlated directly with activity of the bone marrow erythroid series, especially *the* reticulocyte. It is increased in amount in hemolytic anemias and in treated pernicious anemia especially at the height of the reticulocyte response. In iron deficiency there is no significant change It is very low in pernicious anemia in relapse Whether one can interpret the findings in the red cell to indicate the general trend of porphyrin metabolism throughout the body is not clear.

In the inborn disease of porphyrin metabolism, porphyria, large quantities of coproporphyrin and uroporphyrin may appear in the urine and

persons. The amount of food iron offered was for the most part small and perhaps larger amounts might facilitate greater absorption.

Upon entering the mucosal cell the $Fe^{++}OH$ is immediately oxidized to the ferric form in which state it combines with PO_4 radicals in the ratio of nine to one to form a peculiar aggregate. Present in the mucosal cell is a complex protein, apoferritin, of molecular weight 460,000. The ferric hydroxide phosphate aggregates then enter this protein as micelles or clusters, but do not combine chemically. The resultant substance is ferritin which has the same crystalline structure as apoferritin but is amber in color because of the addition of iron. Apoferritin must be made continuously from its precursors because mucosal cells have a tremendous capacity to accept iron, although one is able to detect by chemical methods only small amounts of this protein in the cell. Therefore, one must assume that it is broken down continuously unless ferric hydroxide is present to convert it into stable ferritin. As the body needs more iron the ferritin is broken down to apoferritin and $Fe^{+++}OH$, the latter reduced to $Fe^{++}OH$ in which state it enters the capillaries Thus all the reactions are reversible and a constant state of equilibrium exists between all the elements It is this state that has resulted in the "mucosal block" theory of iron absorption (67). This theory suggests that as the body's need for iron increases, the serum iron falls, ferritin gives up iron to the plasma and more apoferritin is available with which food iron may combine. Therefore, more iron is absorbed The reverse of course would also be true. While this theory explains many of the facts, it fails in isolated instances such as in pyridoxine deficiency in animals or human pernicious anemia (48). In these instances serum iron levels are high, yet the absorption of iron is unimpaired

Another interesting theory which in no way invalidates the "mucosal block" hypothesis is that the relative anoxia conditioned by anemia is responsible for the breakdown of ferritin and the release of $Fe^{++}OH$ into the blood stream. This mechanism could also be responsible for the release of iron from storage. The fact that recently absorbed iron in the mucosal cells is more readily available for hemoglobin synthesis than storage iron could be explained by the greater sensitivity of these cells to anoxia than other cells which have been studied Anoxia might be both a stimulus to red cell formation in the bone marrow and iron availability. Should this theory prove correct, the immediate problem of replacing iron stores in a patient with iron deficiency anemia arises. Presumably as the anemia disappears, iron absorption slows down and iron stores would not be replenished One might wonder if intravenous iron in the form of saccharated oxide of iron should not be utilized to restore these supplies, especially in women, who are constantly losing blood each month during menstruation.

IRON METABOLISM

Many other minerals besides iron may be found within the erythrocyte, but this metal serves an unique and vital function for the mammalian organism. It is an indispensable part of every living cell. Of the total body iron, which represents about 4 5 gm., 60 to 70 per cent is in hemoglobin, 3 to 5 per cent in myoglobin, 0.1 per cent in various enzymes, 0 1 per cent in the serum, and about 15 to 20 per cent in storage in the various cells of the body. *No account can be given for the remaining 5 to 15 per cent.* The utilization of radioactive iron has greatly enhanced our understanding of the absorption, transport, utilization and storage of this element.

That iron is absorbed only in the duodenum has been shown to be untrue. All of the gastro-intestinal mucosa is capable of this function as long as the intestinal contents are at a prescribed pH (70): However, since this optimal pH occurs primarily in the duodenum, a large proportion of the iron is absorbed from this site In the stomach, iron is released from its complex combinations in food by the action of hydrochloric acid (103) Free hydrochloric acid, however, is not absolutely necessary, as persons with achlorhydria may not have any difficulty with iron absorption. Such an example would be a person with pernicious anemia in whom the serum iron is usually elevated. Regardless of this, hydrochloric acid does facilitate the breakdown of bound iron in food into soluble iron salts which are then reduced to ferrous salts in the stomach by many reducing agents such as ascorbic acid, glutathione and sulfhydryl groups of protein digestion products. *These ferrous salts then move into the upper duodenum where the* pH starts to rise. This alkalinity favors the formation of ferrous hydroxide ($Fe^{++}OH$) and ferric hydroxide ($Fe^{+++}OH$). The former, being about *twenty times more* soluble than the latter, enters the mucosal cell (110)

Most of the work on the absorption of food iron prior to 1951 was done utilizing tedious balance studies by Johnston, Frenchman and Boroughs (87) and many other investigators Moore and Dubach (111), however, by ingenious techniques of incorporating radioactive iron into foodstuffs have put the above mechanisms on a sound footing. Although their observations were preliminary and need further study, they showed that. 1) normal human beings as well as iron deficient patients in most cases absorb less than 10 per cent of the iron present in the diet, 2) ascorbic acid definitely increases the absorption of food iron if given with the food sources, cysteine also has this property, 3) *the addition of 0 1 N hydrochloric acid has little* effect on absorption in patients with achlorhydria The most disturbing thing about these findings is that patients with iron deficiency do not absorb food iron any better than normals. This is not true with iron salts, for iron deficient patients do absorb considerably more than do normal

iron oxide. In this instance, the iron is specifically removed by the reticulo-endothelial cells (58, 59).

The incorporation of iron into the protoporphyrin ring to form heme and the subsequent combination with globin is a fascinating chemical reaction about which we know little. The iron atom in the ferrous state has the peculiarity of exerting its coordination valence of six and thereby is able to bind six groups without itself being oxidized. It binds four pyrrole nitrogens in the plane of the ring leaving one group above the plane and one group below. It is postulated that below it is attached to the nitrogen of the imidazole group of histidine in globin. This apparently is a specific attachment which allows O_2 to be accepted by the sixth coordination place above the iron atom. Ferrous heme free in solution is rapidly oxidized to the ferric state, but the attachment of heme to globin allows the O_2 to be accepted and stabilized. Furthermore, the rapidity with which O_2 is combined with heme is approximately 1000 times as great as its dissociation from heme. In the case of CO it is 160,000 times as great.

This particular linkage of iron in heme to the globin also facilitates the so-called isohydric transport of CO_2 in the blood. When O_2 combines with the hemoglobin a proton (H^+) tends to dissociate off, whereas the reverse is true when O_2 is liberated from oxyhemoglobin. The phenomenon of hemoglobin becoming more alkaline with deoxygenation and under these conditions accepting the CO_2 is called the "Bohr" effect. This proton (H^+) is probably lost from the imidazole group of histidine.

One cannot leave iron metabolism without at least mention of the role of copper Fortunately copper is distributed so ubiquitously throughout nature that deficiencies do not exist in adults, and only rarely in infants. However, Wintrobe, Cartwright, Lahey and Gubler (188) studying copper deficiency in swine produced an anemia very similar to that seen in iron deficiency Their preliminary conclusions were that copper either facilitated the absorption of iron from the intestinal tract or was instrumental in releasing iron from its stores—perhaps both The low serum iron level in copper deficiency seemed good evidence against the previous theory that copper was necessary for the incorporation of iron into hemoglobin

ABERRATIONS IN HEMOGLOBIN METABOLISM

The usual forms of hemoglobin, oxyhemoglobin and reduced hemoglobin, have been discussed Other forms of hemoglobin exist which if present in high enough concentrations are capable of producing clinical symptoms of anoxia. These abnormal pigments are carboxyhemoglobin, methemoglobin and sulfhemoglobin which together comprise between two and 12 per cent of the total circulating hemoglobin in normal persons. Whereas five gm per cent of reduced hemoglobin are necessary to produce clinical cyanosis,

Iron in the ferrous state diffuses into the blood stream, is immediately re-oxidized to the ferric form and combines with a protein called "siderophilin", "transferrin" or better "iron binding globulin". This combination requires bicarbonate ion for reasons not clearly understood. This protein is a member of the beta$_1$ globulin fraction with a molecular weight of 90,000. *Each mole of "siderophilin" can carry two atoms of ferric iron.*

This globulin, which is in Cohn's Fraction IV-7, is found in the serum in amounts of 0.24 to 0 28 mg. per cent under normal conditions and is capable of combining with 300 to 359 micrograms of iron per 100 cc. However, normally it is only about one-third saturated, so that the normal serum iron is anywhere from 50 to 180 micrograms per 100 cc Therefore, *there are two more quantitative factors to measure in studying iron metabolism in the patient.* The iron binding capacity of the serum is decreased in infections, uremia, carcinoma, pernicious anemia and cirrhosis. It is increased in iron deficiency, pregnancy, acute blood loss and infectious hepatitis The serum iron is decreased of course in iron deficiency but also in infection, whereas it is increased in pernicious anemia, hemolysis and hypoplastic anemias. *However, unfortunately, no practical significance can be assigned to the serum binding capacity in any anemia yet studied.*

Iron, combined with "siderophilin", is transported either to the bone marrow for immediate utilization as in iron deficiency anemia or to the tissues for storage. Here the iron is released, apparently reduced again to the ferrous hydroxide form and incorporated into hemoglobin or in the case of storage diffuses into the cell and recombines with apoferritin to form ferritin. It is in this form that most of the iron is stored, but it may also be stored as hemosiderin which is assumed to be ferritin with many more molecules of iron within it which transform ferritin from an ordinary invisible molecule to the large readily visualizable hemosiderin deposit. Furthermore there is some evidence that some iron may be stored in the ionizable form as $Fe^{+++}OH$ which would be readily available to the organism.

The normal iron turnover in the human body approximates 25 mg per day. About 97 per cent of this is endogenous iron derived from the body's pool of red cells which undergo destruction The porphyrin ring is broken, and iron split from bilirubin whence it travels via the iron binding globulin to the bone marrow for resynthesis of hemoglobin (58, 59).

The sites of storage are primarily the liver and spleen, but other organs are involved. The peri-portal parenchymal cells of the liver, the reticulo-endothelial cells of the liver and spleen and the macrophages throughout the body are favorite sites. Ordinarily, all of the iron gets to storage iron through the mechanisms outlined above However, the iron binding capacity of the serum may be exceeded by injecting intravenous saccharated

Before concluding this section mention should be made of three abnormal forms of hemoglobin about which information has appeared recently, namely, fetal, sickle cell, and "Type III" hemoglobin. The first is thought to differ from adult hemoglobin in many of its physical properties, including crystallized state, solubility, increased resistance to alkali, electrophoretic mobility, as well as in its amino acid content. The transition from the fetal to the adult form probably occurs during the last few weeks of intra-uterine existence but traces of fetal hemoglobin may be found as long as two years after birth.

Singer, Chernoff and Singer (151) have recently studied various forms of hemoglobin and have described one which resists denaturation by alkali as compared to normal hemoglobin. In this respect it resembles fetal hemoglobin Furthermore, they have found this type of hemoglobin routinely in patients with sickle cell anemia, but not in those with sickle cell trait "F" hemoglobin, as this particular hemoglobin was called, was found in the blood of some patients with thalassemia, hereditary spherocytosis, chronic agenerative anemia, pernicious anemia, leukemia and myelophthistic anemia. However, in some of these diseases it was possible to differentiate this abnormal hemoglobin from true fetal hemoglobin and the conclusions were that it is fetal-like in nature. In other instances it appeared to be indistinguishable from true fetal hemoglobin. The relationship of this abnormal form to the anemia per se has not been clarified as yet.

Not long after Singer's article appeared, Kaplan, Zuelzer and Neel (90) described a hemoglobin which differed from normal and sickle cell hemoglobin in its electrophoretic mobility. This abnormal hemoglobin, tentatively called "Type III", was found in the blood of patients exhibiting mild evidences of hemolytic disease, sickling phenomena and splenomegaly It was present in the cell along with sickle cell hemoglobin. It apparently is transmitted as a Mendelian dominant and may be present with normal hemoglobin in the cells of an ancestor. A homozygous state in relation to "Type III" hemoglobin has not been recognized. How, if at all, this form of hemoglobin differs from Singer's is not clear.

Pauling, Itano, Singer and Wells (119) have shown that the hemoglobin of patients with sickle cell anemia differs in its electrophoretic mobility from that of normal hemoglobin, the former moving as a positive ion and the latter as a negative ion In a study of patients with sickle cell trait these investigators showed that the red cells contained 23 to 45 per cent of sickle cell hemoglobin, whereas sickle cell anemia cells contained 100 per cent. It has been shown that this abnormal hemoglobin when exposed to low O_2 tensions undergoes a molecular rearrangement which has a strikingly similar appearance to that of a sickle cell erythrocyte (75). Such fascinating studies

only 1.5 gm. per cent of methemoglobin and 0.5 gm. per cent of sulfhemoglobin are required to produce the same degree of cyanosis (189). Carboxyhemoglobin is bright red, rather than dark, so that the characteristic cherry red color is imparted to patients poisoned with CO.

Methemoglobin differs from hemoglobin in that the iron is oxidized from the ferrous to the ferric state. In this state it is unable to carry O_2 and thus is rendered useless for respiration. Normally this form is present in a concentration not greater than 1 7 per cent (98). This amount of methemoglobin is stabilized by the presence in the red cell of an elaborate enzyme system capable of metabolizing glucose to lactic acid As a by-product of this glycolysis, a reduced form of the diphosphopyridine nucleotide (coenzyme I) arises which is capable of reducing the ferric hemoglobin to its original state (66). It can be calculated that the glycolysis of one molecule of glucose can reduce four hemes of methemoglobin. Although this is an efficient method which can handle the ordinary load of methemoglobin, this system is unable to cope with the situation which obtains when the blood is exposed to large amounts of a chemical compound capable of preferentially oxidizing hemoglobin. Thus many compounds, among which are anilin dyes, phenacetin, acetanilid, sulfonamides, various nitrites including nitroglycerin and many others, are capable of producing clinical cyanosis and anoxia in human beings. In addition there is described an "hereditary" form which apparently is due to a defective reducing mechanism, possibly involving a dysfunction of coenzyme I (1).

A powerful reducing substance, such as methylene blue, is valuable therapeutically in reducing the concentration of methemoglobin in the acquired variety, whereas, ascorbic acid performs adequately in the "hereditary" variety.

Sulfhemoglobin, a much rarer form, differs from methemoglobin in that the hemoglobin molecule undergoes a change in structure. Labile sulfur enters the molecule and heme is attached to globin in a different fashion. *It is formed by the reaction of hemoglobin with soluble sufides and hydrogen sulfide* Sulfhemoglobinemia often accompanies methemoglobinemia and has been found in persons taking those chemicals mentioned above Unlike methemoglobin, sulfhemoglobin is not convertible directly to hemoglobin, but must be completely disassociated and destroyed and *new normal* hemoglobin rebuilt

Carboxyhemoglobin represents merely a direct replacement of the O_2 molecule in oxyhemoglobin by the CO molecule. CO has an affinity for hemoglobin 210 times that of O_2 so that toxic amounts can be produced in the blood by inhalation of relatively small amounts of the gas Administration of O_2 and CO_2 mixtures with some form of artificial respiration is the only therapy for this poisoning.

deficiency in and of themselves. Therefore any adult presenting himself with hypochromic anemia must be thoroughly examined both physically and roentgenologically for possible sites of blood loss.

The human body is endowed with an extreme economy of iron. Normally each day a total of approximately one mg. of iron is excreted via urine, bile, sweat, exfoliated epithelial cells, etc., exclusive of menstruation (71). 1.5 mg. of iron on the average (the maximum is three mg.) are absorbed each day, so that the margin of safety is very low. Approximately 0.5 mg. of iron is contained in each one cc. of normal blood. Simple mathematics will indicate how moderate blood loss such as in normal menstruation may seriously deplete the iron stores of the body. Add to this the 500 mg. of iron contributed to the baby during each pregnancy, plus 300 mg. during lactation for one year and one appreciates why hypochromic anemia is the most common anemia in women of the childbearing age. Increased needs during periods of active growth also strain iron stores. Generally a careful history will divulge the cause of the anemia in such cases.

Deficiency in nutritional intake may play a role, but is rare in this country today. Chlorosis, a common disease of adolescent girls around the turn of the century, was undoubtedly due to a combination of poor diet, increased demand of the body for growth at a crucial time, and most important the onset of menstruation with its concomitant loss of iron. This situation led to a severe iron deficiency anemia which produced great morbidity and considerable mortality. The disease is rare at present, probably in large part because of better pediatric care as well as the common use of soluble iron salts. At the present time an average dietary intake of 15 mg. of iron per day is recommended although this varies depending on age and special requirements. This is easily met by most diets. Eggs, meat and dried fruits are particularly good sources.

Absorptive difficulty presents a real challenge to the body's economy, but rarely is it the principal factor in the production of iron deficiency. However, 50 to 60 per cent of adult patients with this type of anemia have histamine refractory achlorhydria. What role ascorbic acid deficiency actually plays in the production of iron deficiency is unclear since this type of anemia is unusual in scurvy and serum iron levels are normal (175). Generally the anemia of adult scurvy is normocytic or slightly macrocytic. Certainly diseases characterized by poor absorptive capacities of the gastrointestinal mucosa, such as sprue and myxedema, may result in a hypochromic anemia, but usually in these diseases deficiencies of other nutritional factors overshadow the iron deficiency, and macrocytic anemias result.

There is another distinct aberration of iron metabolism which results in a human disease, hemochromatosis. In this condition, clinically characterized by the classical triad of bronzing of the skin, diabetes mellitus

are contributing much to our understanding of this particular hemolytic anemia

CLINICAL MANIFESTATIONS OF DERANGEMENTS IN HEMOGLOBIN METABOLISM

In human beings anemia associated with an abnormality in porphyrin metabolism is not recognized, except for the hemolytic anemia of congenital porphyria. Whether the latter is etiologically related to the aberration of porphyrins is not clear at this time.

As with protoporphyrin, defects in globin metabolism have not been directly proved to be responsible for anemia in human beings Deficiency of certain amino acids is capable of producing anemia in animals (17) but because of the complexity of the problem and obvious difficulties inherent in such an experiment isolated amino acid deprivation studies in humans have been few. However, severe animal protein deficiency has been associated with a normocytic or slightly macrocytic anemia in humans. This was especially evident in starved war prisoners. Here again there are multiple factors complicating the problem, but we have been impressed with the rare case of slightly macrocytic anemia associated with a fatty, hypocellular bone marrow, malnutrition and hypoproteinemia These anemias do not respond to any specific hematopoietic factor, but do respond slowly to general dietary improvement, especially increased protein ingestion Such an anemia may well represent a defect in globin metabolism, but such a direct relationship has not been proved

Abnormalities of iron metabolism account for the large proportion of human afflictions attributable to aberrations in hemoglobin manufacture. A few important points will be mentioned

Lack of iron in the erythrocyte results in a specific decrease in the concentration of hemoglobin in the average cell (MCHC) called hypochromia. This deficiency of iron may be the result of some primary abnormality in iron metabolism or some defect in iron utilization conditioned by another disease such as anemias associated with infection, uremia and malignancy. In these conditions the ability of the serum to combine with iron is decreased However, there is no direct proof of any etiological relationship between iron binding capacity and anemia, and indeed, absolute hypochromia is rare in these anemias Furthermore, the administration of iron in no way affects the hemoglobin level in these instances

Primary disturbances in iron metabolism are responsible for the great majority of hypochromic anemias in humans Of these the most result from loss of iron from the body via bleeding. Naturally all of the factors reviewed earlier as being important such as pH of the gastric juice, reducing substances in food, integrity of the intestinal mucosa, etc. all may play a contributing role but are probably not of sufficient import to produce iron

discussed in this monograph. Ribonucleic acid seems to be essential for the synthesis of protein, a prerequisite to cell growth. Desoxyribonucleic acid is necessary for cell division. The synthesis of both types of nucleic acid depends upon the availability of folic acid, ascorbic acid and vitamin B_{12}. Folic acid, in the presence of ascorbic acid, may be converted into an active catalyst which facilitates the transfer of single carbon units and helps to form the purine and probably the pyrimidine rings from simple compounds like glycine and formate. Vitamin B_{12} also catalyzes methylation reactions and seems to be involved in the formation of ribosides and ribotides of the purines and pyrimidines. When any stage of the very complicated chain reaction catalyzed by these and other unknown coenzymes is disturbed, megaloblastic anemia may occur. Consideration of these reactions helps to explain the genesis of pernicious anemia and related macrocytic megaloblastic anemias.

Chemical reactions, equally as complicated, are necessary for the formation of hemoglobin which takes place after ribonucleic acid has begun to disappear. We have tried to trace these reactions which involve the formation of the pyrrol ring from simple compounds like glycine and acetate, the formation of the more complex porphyrins which contain four pyrrol rings, the inclusion of coordinately linked ferrous iron to form heme, and the conjugation of heme with globin. Pyridoxine may be necessary for the formation of the pyrrol ring, possibly because of its codecarboxylase function and folic acid may facilitate the transfer of carbon into the final protoporphyrin molecule. Any abnormality which may develop in this complicated reaction may cause microcytic hypochromic anemia. Insufficient supplies of iron to complete the formation of heme is the most common cause.

We have attempted to discuss all the abnormalities known or suspected which may possibly affect the chemistry and physiology of erythrocyte development and cause anemia in human beings. Much of this work is purely speculative. The theories which have been reviewed in this monograph and the studies done to test their validity have led to a rapidly expanding body of knowledge concerning the chemical reactions which occur during the growth and maturation of erythroblasts to erythrocytes. Even more important than their relationship to the occurrence of anemia, is their bearing on fundamental problems of cell growth. The reactions described are essential for the growth of all bone marrow cells and probably for the growth of benign and malignant tumors as well. When the chemistry and physiology of growth, division and maturation of normal cells are known, the basic mechanisms of malignant or unfettered growth may not be too difficult to discover, and once discovered, therapeutic measures to meet the problem of control of malignant tumors will be close at hand.

and cirrhosis, iron is deposited in the tissues in excessive amounts as hemosiderin, giving rise to fibrosis and cellular degeneration There is another non-iron pigment present, "hemofuscin", the significance of which is unclear. Granick (67) looks upon hemochromatosis as a constitutional inadequacy of the "mucosal block" due to a slightly greater reducing tendency of the cell and that over a period of years enough iron is absorbed and stored, without any loss occuring, to produce the clinical picture of hemochromatosis. Increased absorption of radioactive iron in this disease was demonstrated first by Dubach, Callender and Moore (48). The disease has been described in patients who had received many transfusions over a period of years and presumably had stored excessive amounts of iron from the breakdown of the infused red cells (144). The possible dangers of excessive administration of intravenous iron are obvious Indiscriminate use of such preparations might well lead to the development of hemochromatosis. At the present time the dividing line between hemosiderosis and hemochromatosis is an arbitrary one and may depend on time more than quantity of iron. The very logical choice of therapy in this disease, repeated phlebotomy, has been suggested by Davis and Arrowsmith (38). They reported definite clinical, laboratory and pathological evidence of improvement in patients following repeated bleedings without development of anemia or any other untoward reaction.

Fortunately the therapy of iron deficiency is simple. It consists of the administration of soluble ferrous salts such as ferrous sulfate or ferrous gluconate. Optimal dosage has been set at 1.2 gm. per day in three divided doses Additional hydrochloric acid or reducing substances are unnecessary. Intravenous iron in the form of saccharated iron oxide given in carefully calculated doses may be needed in the rare person unable to take oral iron for one reason or another, in the patient with ulcerative colitis where oral iron may irritate the already inflamed gut, or in the iron deficient pregnant woman who is diagnosed or seen very late in preganancy and the physician wishes to replenish the iron stores of fetus and mother before delivery The routine use of intravenous iron in ordinary iron deficiency should be frowned upon as an useless expense to the patient as well as a potential hazard if the dose is not carefully controlled

RESUMÉ

The erythrocyte, composed of protein, remnants of nucleic acid, hemoglobin, phospholipids and related compounds, enzymes and coenzymes, and inorganic elements develops from the erythroblast because of many complicated chemical reactions. What is known of these reactions and the types of anemia which occur when abnormalities develop in them have been

M.: Experimental production of a nutritional macrocytic anemia in swine Blood, 4: 301, 1949.

19 Cartwright, G E, Tatting, B S, Robinson, J, Fellows, N M, Gunn, F D. and Wintrobe, M. M.: Hematologic manifestations of vitamin B_{12} deficiency in swine. Blood, 6: 867, 1951.

20. Case, R A M.: Siderocytes in hemolytic diseases a new index of severity and progress J Path. & Bact., 57: 271, 1945

21. Caspersson, T and Schultz, J.: Ribonucleic acids in both nucleus and cytoplasm and the function of the nucleolus Proc. Nat Acad. Sci, Wash, 26: 507, 1940

22 Caspersson, T. and Santesson, L.. A review of the mechanisms of protein and nucleic acid metabolism in normal cells. Acta Radiol, Suppl #46, 1942

23 Caspersson, T.: Relations between nucleic acid and protein synthesis Symposia, Soc. Exp Biol, I, p 127. Cambridge Univ Press Cambridge, 1947

24 Castle, W. B.: Observations on the etiologic relationship of achylia gastrica to pernicious anemia. I The effect of the administration to patients with pernicious anemia of the contents of the normal human stomach recovered after the ingestion of beef muscle. Am J. Med Sci, 178: 748, 1929

25 Clemmesen, J, Espersen, T. and Plum, C M In vitro study of bone marrow III Erythropoiesis in vitro of spinal marrow from cases of pernicious anemia and lymphatic leucosis under therapy. Blood, 3: 155, 1948

26 Cooley, G, Ellis, B, Petrow, V., Beaven, G H, Holiday, E. R and Johnson, E A. The chemistry of some antipernicious anemia factors VII Some transformations of vitamin B_{12b}. J. Pharm & Pharmacol, 3: 271, 1951

27. Dacie, J. V and White, J. C.. Erythropoiesis with particular reference to its study by biopsy of human bone marrow, a review J Clin Path, 2: 1, 1949

28 Darby, W J. and Jones, E: Treatment of sprue with synthetic L casei factor (folic acid, vitamin M). Proc Soc. Exp Biol & Med, 60: 259, 1945

29. Daughaday, W. H., Williams, R H and Daland, G A The effect of endocrinopathies on the blood Blood, 3: 1342, 1948

30 Davidson, C S, Murphy, J. C., Watson, R J and Castle, W B Comparison of effects of massive blood transfusions and of liver extract in pernicious anemia J Clin Invest, 25: 858, 1946

31 Davidson, J. N.: Some factors influencing nucleic acid content of cells and tissues Symposium on Quantitative Biology (Biol Lab, Cold Springs Harbor), 12: 50, 1947.

32 Davidson, J N, Leslie, I. and White, J C The nucleic acid content of the cell Lancet, 1: 1287, 1951.

33 Davidson, L S. P.: The basophilic substance of the erythrocyte Edinburgh Med J, 37: 425, 1930

34 Davidson, L S. P, Girdwood, R H and Clark, J R *Pernicious anemia of pregnancy and the puerperium* Br Med J, 1: 819, 1948

35 Davidson, L S P. and Lond, F R S Refractory megaloblastic anemia Blood, 3: 107, 1948

36 Davis, B D and Mingioli, E S Mutants of Escherichia coli requiring methionine or vitamin B_{12} J Bact, 60: 17, 1950

37 Davis, L. J and Brown, A The erythropoietic activity of choline chloride in megaloblastic anemias Blood, 2: 407, 1947

38 Davis, W D. and Arrowsmith, W R The effect of repeated bleeding in hemachromatosis J. Lab. & Clin Med, 36: 814, 1950

REFERENCES

1 BARCOFT, H., GIBSON, Q H, HARRISON, D. C. AND MCMURRAY, J.. Familial idiopathic methemoglobinemia and its treatment with ascorbic acid Clin Sci, 5: 145, 1945
2 BARCOFT, J The significance of hemoglobin in submammalian forms of life Physiol Rev, 4: 329, 1924.
3 BEARD, M F AND MCILVANIE, S K Vitamin B_{12} as a chemotherapeutic agent in pernicious anemia. Proc. Third Internat Cong, Internat. Soc Hematology, p 34 Grune and Stratton, New York, 1951.
4 BEAVEN, G H, HOLIDAY, E R., JOHNSON, E A, ELLIS, B AND PETROW, V. The chemistry of anti-pernicious anemia factors. J. Pharm. & Pharmacol, 2: 944, 1950.
5 BERK, L, CASTLE, W B, WELCH, A D, HEINLE, R. W., ANKER, R. AND EPSTEIN, M.: Observations on the etiologic relationship of achylia gastrica to pernicious anemia X Activity of vitamin B_{12} as food (extrinsic) factor. New Eng J. Med, 237: 911, 1948
6 BETHELL, F H, MEYERS, M C, ANDREWS, G A, SWENDSEID, M E, BIRD, O D AND BROWN, R A Metabolic function of pteroylglutamic acid and its hexaglutamyl conjugate I Hematologic and urinary excretion studies on patients with macrocytic anemia J Lab & Clin Med, 32: 3, 1947
7 BETHELL, F H, MEYERS, M C AND NELIGH, R B Vitamin B_{12} in pernicious anemia and puerperal macrocytic anemia J Lab & Clin Med, 33: 1477, 1948
8 BIRD, R M, CLEMENTS, J A AND BECKER, L M Metabolism of leukemic cells in vitro Fed Proc, 9: 11, 1950
9 BLOOM, M L AND WISLOCKI, G B The localization of lipids in human blood and bone marrow cells Blood, 5: 79, 1950
10 BOSTRÖM, L Are non nucleated erythrocytes formed by budding off of cytoplasm from normoblasts? Acta Med Scandinav, 131: 303, 1948
11 BROCKMAN, J A, JR, ROTH, B, BROQUIST, H P, HULTQUIST, M E, SMITH, J M, JR, FAHRENBACH, M J, COSULICH, D B, PARKER, R P, STOKSTAD, E L R AND JUKES, T H Synthesis and isolation of a crystalline substance with the properties of a new B vitamin J Am Chem Soc, 72: 4325, 1950
12 BROWN, G B, ROLL, P M, PLENTL, A A AND CAVALIERI, L F The utilization of adenine for nucleic acid synthesis and as a precursor of guanine J. Biol Chem, 172: 469, 1948
13 BURCHENAL, J H, JOHNSTON, S F AND WARING, G B Mechanisms of amethopterin resistance in leukemia I Effects of weak folic acid antagonists on mouse leukemias Proc Soc Exp Biol & Med, 78: 348, 1951
14 BURCHENAL, J H, WEBBER, L F AND JOHNSTON, S F Mechanism of amethopterin resistance in leukemia II Effect of cortisone on sensitive and resistant mouse leukemias Proc Soc Exp Biol & Med, 78 352, 1951
15 CALLENDER, S T AND LAJTHA, L G On the nature of Castle's hematopoietic factor Blood, 6: 1234, 1951
16 CARTWRIGHT, G. E, WINTROBE, M M AND HUMPHREYS, S Studies on aemia in swine due to pyridoxine deficiency, together with data on phenylhydrazine anemia J Biol Chem, 153: 171, 1944
17 CARTWRIGHT, G E Dietary factors concerned in erythropoiesis Blood, 2: 111, 1947
18 CARTWRIGHT, G E, TATTING, B S, ASHENBRUCKER, H. AND WINTROBE, M

60 GABUZDA, G. J., JR, PHILLIPS, G. B, SHILLING, R. F AND DAVIDSON, C S Metabolism of pteroylglutamic acid and citrovorum factor in human scurvy J Clin Invest. Proc, 30: 639, 1951.
61. GILLIS, M B. AND NORRIS, L. C.: Effect of the animal protein factor on the requirement for methylating compounds J Biol. Chem, 179: 487, 1949
62. GINSBERG, V., WATSON, J. AND LICHTMAN, H.: Megaloblastic anemia of pregnancy. response to pteroylglutamic acid after failure to respond to liver extract and vitamin B_{12}. J Lab & Clin Med., 36: 238, 1950
63 GORDON, A. S. AND CHARIPPER, H A: The endocrine system and hematopoiesis Ann. N. Y Acad. Sci, 48: 615, 1947.
64 GOVAN, C. D, JR AND GORDON, H. H The effect of pteroylglutamic acid on the aromatic acid metabolism of premature infants Science, 109: 332, 1949
65 GRANICK, S AND GILDER, H.: Distribution, structure and properties of the tetrapyrroles. Adv. Enzymol, 7: 305, 1947
66 GRANICK, S: The chemistry and functioning of the mammalian erythrocyte Blood, 4: 404, 1949.
67. GRANICK, S: Iron metabolism and hemachromatosis Bull N Y Acad Med, 25: 403, 1949
68 GREENBERG, G R Mechanism of biosynthesis of purine Fed Proc, 9: 179, 1950
69 GREENBERG, G. R.: De novo synthesis of hypoxanthine via inosine-5-phosphate and inosine. J Biol Chem, 190: 611, 1951
70 HAHN, P F, BALE, W F., LAWRENCE, E O AND WHIPPLE, G H Radioactive iron and its metabolism in anemia: its absorption, transportation and utilization J. Exp Med, 69: 739, 1939.
71 HAHN, P. F., BALE, W. F, HETTIG, R. A, KAMEN, M D AND WHIPPLE, G H Radioactive iron and its excretion in urine, bile and feces J Exp Med, 70: 443, 1939.
72 HAMMARSTEN, E. AND HEVESY, G.: Rate of renewal of ribo- and desoxyribonucleic acid Acta Physiol. Scandinav, 11: 335, 1946
73 HAMMARSTEN, E, REICHARD, P AND SALUSTE, E Pyrimidine nucleosides as precursors of pyrimidines and polynucleotides J Biol Chem, 183: 105, 1950
74 HANSEN-PRUSS, O. C.: Relapse of patients with pernicious anemia receiving folic acid Am J Med Sci, 214: 465, 1947
75 HARRIS, J. W.: Studies on the destruction of red blood cells VIII Molecular orientation in sickle cell hemoglobin solutions Proc Soc Exp Biol & Med, 75: 197, 1950
76 HAUSMANN, K: Haematopoietic effect of thymidine in pernicious anemia Lancet, 1: 329, 1951.
77. HEATH, C W AND DALAND, G. A The life of reticulocytes, experiments on their maturation. Arch Int. Med, 46: 533, 1930
78 HEINLE, R W. AND WELCH, A D: Folic acid in pernicious anemia Failure to prevent neurologic relapse J A M A, 133: 739, 1947
79 HEINLE, R W., WELCH, A D, GEORGE, W L, EPSTEIN, M AND PRICHARD, J A Effect of extrinsic factor, liver extract and folic acid on induced macrocytic anemia of swine J. Lab & Clin Med, 32: 1398, 1947
80 HEINLE, R W, WELCH, A D AND SHORR, H L Interrelation of pteroylglutamic acid and vitamin B_{12} in induced anemia in swine J Lab & Clin Med, 34: 1763, 1949
81 HOLLY, R. G.: Megaloblastic anemia in pregnancy Remission following com-

39. DEMEREC, M., Editor. nucleic acids and nucleoproteins. Cold Springs Harbor Symposia, Quant. Biol., 12: 1, 1947
40. DIETRICH, L S., NICHOL, C. A., MONSON, W. J AND ELVEHJEM, C. A : Observations on the interrelation of vitamin B_{12}, folic acid and vitamin C in the chick J. Biol. Chem , 181: 915, 1949
41. DIETRICH, L S., MONSON, W J. AND ELVEHJEM, C. A · Observations on a relationship between vitamin B_{12}, folic acid and the citrovorum factor. Proc. Soc. Exp. Biol & Med., 77: 93, 1951.
42 DIETZ, A. A Chemical composition of normal bone marrow. Arch Biochem , 23: 211, 1949.
43 DIETZ, A A. AND STEINBERG, B Chemical studies of normal and leukemic bone marrow Blood, 6: 175, 1951
44 DILLE, R. S AND WATKINS, C H.: Methods for the determination of plasma catalase and the values obtained in normal adults J Lab. & Clin Med , 33: 480, 1948.
45 DINNING, J S , PAYNE, L D. AND DAY, P. L · The requirements of rats for methyl groups and vitamin B_{12} in the production of leucocytes Arch Biochem , 27: 467, 1950
46 DOAN, C A. Current views on the origin and maturation of the cells of blood J Lab & Clin Med , 17: 887, 1932
47 DOWNEY, H Handbook of Hematology Paul B Hoeber, Inc , New York, 1938.
48. DUBACH, R , CALLENDER, S. T E. AND MOORE, C. V Studies in iron transportation metabolism VI Absorption of radioactive iron in patients with fever and with anemias of varied etiology Blood, 3: 526, 1948
49 DUBNOFF, J W *The effect of B_{12} concentrates on the reduction of S—S groups* Arch Biochem , 27: 466, 1950.
50. DUVIGNEAUD, V., RESSLER, G AND RACHELE, J. R The biological synthesis of "labile methyl groups". Science, 112: 267, 1950
51 ELLISON, R R , WOLFE, S , LICHTMAN, H , GINSBERG, V. AND WATSON, J : Effect of citrovorum factor in pernicious anemia Proc Soc Exp Biol & Med , 76: 366, 1951
52. ELWYN, D AND SPRINSON, D B The relation of folic acid to the metabolism of serine J. Biol Chem , 184: 475, 1950
53. EMERSON, G AND FOLKERS, K Water soluble vitamins Ann Rev Biochem , 20: 559, 1951
54 ENGELBRETH-HOLM, J AND PLUM, C M Production of stippled erythrocytes in vitro Nature, 166: 990, 1950
55 EVANS, J. D AND BIRD, R M Metabolism of rabbit bone narrow in vitro in Ringer-bicarbonate medium containing no added glucose J Biol Chem , 181: 357, 1949
56 FALCONER, E H The clinical significance of punctate basophilia in the erythrocyte Ann Int Med , 12: 1429, 1939
57. FERTMAN, M H AND DOAN, C A Irreversible toxic "inclusion body" anemia, rarely recognized syndrome, clinical and experimental studies Blood, 3: 349. 1948

59. FINCH, C. A, [illegible] KINS, D., FINCH, S AND FLUHARTY, R G Iron metabolism the pathophysiology of iron storage. Blood, 5: 983, 1950

megaloblastic anemia in infancy, an interrelationship between pteroylglutamic acid and ascorbic acid. Am. J. Dis. Child., 80: 191, 1950.
103. METTIER, S R. AND MINOT, G. R.: Effect of iron on blood formation as influenced by changing the acidity of gastroduodenal contents in certain cases of anemia Am. J. Med. Sci, 181: 25, 1931.
104 MEYER, L. M., SAWITSKY, A, COHEN, B S., KRIM, M. AND FADEM, R: Oral treatment of pernicious anemia with vitamin B_{12}. Am. J. Med. Sci, 220: 604, 1950.
105. MEYER, L. M., BRAHIN, C. M. AND SAWITSKY, A.: Treatment of pernicious anemia with citrovorum factor. Proc. Soc Exp. Biol. & Med., 76: 86, 1951.
106. MEYER, O. O., STEWART, G E, THEWLIS, E. W. AND RUSCH, H. P.: Hypophysis and hematopoiesis Folia Hemat, 57: 99, 1937.
107 MICHELS, N. A: The erythrocyte, critical review of its normal and pathologic morphology and physiology with data on normal red cell count and technic. Haematologica, 2: 101, 1931.
108. MINOT, G. R AND MURPHY, W. P.: Treatment of pernicious anemia by a special diet J. A. M. A, 87: 470, 1926
109 MITCHELL, H. K. AND HOULAHAN, M. B.: Investigations on the biosynthesis of pyrimidine nucleosides in neurospora Fed Proc, 6: 506, 1947.
110 MOORE, C. V, DUBACH, R, MINICH, V. AND ROBERTS, H. K. Absorption of ferrous and ferric radioactive iron by human subjects and by dogs. J Clin. Invest, 23: 755, 1944
111 MOORE, C. V. AND DUBACH, R.: Observations on the absorption of iron from foods tagged with radioiron. Trans. Assoc. Am Phys, 64: 245, 1951.
112 MORGANS, M. E, RIMINGTON, C AND WHITTAKER, N. Folic acid in megaloblastic anemia after total gastrectomy; report of case. Lancet, 2: 128, 1947.
113 MUELLER, J F, HAWKINS, V. R AND VILTER, R W. Liver extract refractory megaloblastic anemia. Blood, 4: 1117, 1949
114 MUELLER, J. F. AND VILTER, R W.: Pyridoxine deficiency in human beings induced with desoxypyridoxine. J. Clin. Invest, 29: 193, 1950
115 NEUBERGER, A, MUIR, H. M. AND GRAY, C H: Biosynthesis of porphyrins and congenital porphyria Nature, 165: 948, 1950; Biochem. J., 47: 97, 1950.
116 NICHOL, C A AND WELCH, A D · Synthesis of citrovorum factor from folic acid by liver slices augmentation by ascorbic acid Proc Soc. Exp Biol. & Med, 74: 52, 1950
117 NICHOL, C A AND WELCH, A D · On the method of action of aminopterin Proc Soc Exp. Biol & Med, 74: 402, 1950
118 OWREN, P A Congenital hemolytic jaundice The pathogenesis of the "hemolytic crisis" Blood, 3: 231, 1948
119 PAULING, L, ITANO, H A, SINGER, S S AND WELLS, I. C. Sickle cell anemia, a molecular disease Science, 110: 543, 1949; J Biol. Chem, 187: 221, 1950
120 PEARLMAN, M D AND LIMZARZI, L R Correlation studies of basophilic aggregation and reticulocytes in various clinical conditions Am J. Clin Path, 8: 608, 1938
121 PETERING, H G AND DELOR, R A Inability of thymine and adenine to substitute for pteroylglutamic acid in the folic acid deficient rat Science, 110: 185, 1949
122 PLAUT, G W E, BETHEIL, J J. AND LARDY, H A.· The relationship of folic acid to formate metabolism in the rat J Biol Chem, 184: 795, 1950

bined therapy with ascorbic acid and vitamin B_{12} Proc. Soc. Exp Biol Med., 78: 238, 1951.

82. HORRIGAN, D., JARROLD, T. AND VILTER, R. W.: Direct action of vitamin B_{12} upon human bone marrow. J. Clin. Invest, 30: 31, 1951.
83. HOWELL, W. H.: *The life history of the formed elements of the blood: especially the red corpuscles* J. Morphology, 4: 57, 1890.
84. ISAACS, R: The refractive granule red blood corpuscle: its behavior and significance Anat. Rec, 29: 299, 1925.
85. JARROLD, T., HORRIGAN, D., THOMPSON, C. AND VILTER, R. W: The hematologic effect of folinic acid (citrovorum factor) in persons with pernicious anemia. Science, 113: 688, 1951.
86. JENNINGS, G. H. AND GLAZEBROOK, A. J: A comparison of clinical and blood pictures in adult scurvy. Br. Med J., 2: 784, 1938
87. JOHNSTON, F. A, FRENCHMAN, R. AND BOROUGHS, E D.: The iron metabolism of young women on two levels of intake. J. Nutrition, 38: 479, 1949.
88. JUKES, T. H. AND STOKSTAD, E. L. R The role of vitamin B_{12} in metabolic processes Vitamins and Hormones, 9: 1. Academic Press, Inc., New York, 1951
89. KACZKA, E. A., WOLF, D. E, KUEHL, F. A., JR. AND FOLKERS, K.: Vitamin B_{12}, reactions of cyano-cobalamin and related compounds Science, 112: 354, 1950
90. KAPLAN, E, ZUELZER, W. W. AND NEEL, J. V.: A new inherited abnormality of hemoglobin and its interaction with sickle cell hemoglobin. Blood, 6: 1240, 1951.
91. KATO, K.. Monophyletic scheme of blood cell formation for clinical and laboratory reference J. Lab. & Clin Med, 20: 1243, 1935.
92. KERESZTESY, J. C AND SILVERMAN, M: Crystalline citrovorum factor from liver. J. Am Chem Soc., 73: 5510, 1951.
93. KERPPOLA, W.. Observations on the phosphatase content of blood and bone marrow cells in normal and pathologic hematopoiesis Blood, 6: 454, 1951.
94. KEY, J. A: Studies of erythrocytes with special reference to reticulum, polychromatophilia and mitochondria. Arch Int Med., 28: 511, 1921
95. KLEIN, J. R AND KOHN, H I. Synthesis of flavin-adenine dinucleotide from riboflavin by human blood cells in vitro and in vivo. J. Biol. Chem, 136: 177, 1940.
96. KOHN, H I AND KLEIN, J. R. The synthesis of cozymase and of factor V from nicotinic acid by the human erythrocyte in vitro and in vivo. J Biol Chem, 130: 1, 1939
97. LAJTHA, L. G An inhibitory factor in pernicious anemia serum Clin Sci, 9: 287, 1950

[illegible] -ter-

[illegible] ;cin

in pernicious anemia [illegible]

100. LUHBY, A. L. AND WHEELER, W E Megaloblastic anemia of infancy II Failure of response to vitamin B_{12} and the metabolic role of folic acid and vitamin C. Ohio State Univ. Health Center J, 3: 1, 1949.
101. MAY, C D, SUNDBERG, D AND SCHAAR, F Comparison of effects of folic acid and folinic acid in experimental megaloblastic anemia J Lab & Clin. Med, 36: 963, 1950.
102. MAY, C. D, NELSON, E. N, LOWE, C U. AND SALMON, R. J. Pathogenesis of

146 SHEMIN, D AND WITTENBERG, J : The mechanism of porphyrin formation The role of the tricarboxylic acid cycles, J Biol. Chem , 192: 315, 1951
147 SHIVE, W , RAVEL, J. M. AND EAKIN, R. E.: An interrelationship of thymidine and vitamin B_{12}. J Am. Chem Soc , 70: 2614, 1948
148 SHIVE, W.: The functions of B vitamins in the biosynthesis of purines and pyrimidines Vitamins and Hormones, 9: 75 Academic Press, Inc , New York, 1951.
149 SHORB, M S · *Activity of vitamin B_{12} for growth of Lactobacillus lactis Science,* 107: 397, 1948
150 SIEBENTHAL, B. J., Megaloblastic anemia of infancy. J. Ped , 32: 188, 1948
151 SINGER, K , CHERNOFF, A. I AND SINGER, L., Studies on abnormal hemoglobins I Their demonstration in sickle cell anemia and other hematologic disorders by means of alkali denaturation. Blood, 6: 413, 1951, ibid, 6: 429, 1951
152 SMITH, E L · Purification of antipernicious anemia factors from liver Nature, 161: 638, 1948
153 SMITH, E L · The vitamin B_{12} group of factors Br Med J , 1: 151, 1951
154 SNELL, E E AND MITCHELL, H. K · Purine and pyrimidine bases as growth substances for lactic acid bacteria Proc Nat Acad Sci U S , 27: 1, 1941
155 SPIES, T D , VILTER, C. F , KOCH, M B. AND CALDWELL, M H Observations on the antianemia properties of synthetic folic acid South Med J , 38: 707, 1945
156 SPIES, T. D , FROMMEYER, W. B , JR , VILTER, C F AND ENGLISH, A Antianemic *properties of thymine Blood,* 1: 185, 1946
157. SPIES, T D , LOPEZ, G G, MILANES, F., STONE, R E , TOCA, R L AND ARAMBURU, T.· Treatment of nutritional macrocytic anemia with synthetic folic acid Lancet, 1: 239, 1948
158 *SPIES*, T. D. AND SUAREZ, R. M : Response of tropical sprue to vitamin B_{12} Blood, 3: 1213, 1948
159 SPIES, T. D , STONE, R. E. AND ARAMBURU, T Observations on the antianemic properties of vitamin B_{12} South. Med J , 41: 522, 1948
160 SPIES, T. D , GARCIA-LOPEZ, G , MILANES, F , LOPEZ-TOCA, R AND CULVER, B · Observations on the hematopoietic response of persons with tropical sprue to vitamin B_{12} South Med J , 41: 523, 1948
161 STEDMAN, E AND STEDMAN, E The cytological interpretation of the Feulgen reaction Biochem J , 47: 508, 1950
162 STEKOL, J. A AND WEISS, K Vitamin B_{12} and growth of rats on diets free of methionine and choline J Biol Chem , 186: 343, 1950
163 STOKES, J. L. Substitution of thymine for "folic acid" in the nutrition of the of the lactic acid bacteria J Bact , 48: 201, 1944
164 STONE, R E AND SPIES, T D Adenine its failure to stimulate hematopoiesis or to produce pellagra in a case of pernicious anemia Am J Med Sci , 215: 411, 1948
165 THOMA, K . The detection of ribonucleinic acids in erythrocytes Klin Wchnschr , 28· 215, 1950
166 THOMPSON, R B Addisonian premicious anemia Confirmatory evidence of a factor inhibiting erythropoiesis Clin Sci , 9: 281, 1950
167. THORELL, B Studies on the formation of cellular substances during blood cell production Acta Med Scandinav , Suppl 200, 1, 1947
168 TINSLEY, J. C., JR , MOORE, C V , DUBACH, R , MINNICH, V AND GRINSTEIN, M : Role of oxygen in regulation of erythropoiesis Depression of rate of de-

123. Plentl, A. A. and Schoenheimer, R.: Studies in the metabolism of purines and pyrimidines by means of isotopic nitrogen. J. Biol. Chem., 153: 203, 1944
124. Ponder, E.: Red cell cytochemistry and architecture. Ann N. Y. Acad. Sci., 48: 579, 1947.
125 Porter, R. and Sanger, F.: The free amino groups of hemoglobin. Biochem J., 42: 287, 1948
126. Prusoff, W. H., Tepley, L J. and King, C. G: The influence of pteroylglutamic acid on nucleic acid synthesis in Lactobacillus casei. J Biol Chem, 176: 1309, 1948.
127. Rebuck, J. W.. The function of the white blood cells Am. J. Clin Path, 17: 614, 1947.
128 Reisner, E. H., Jr and West, R.: Effect of thymine desoxyriboside (thymidine) on human pernicious anemia Proc Soc. Exp. Biol. & Med, 71: 651, 1949
129 Reisner, E. H, Jr. and Korson, R: Microspectrophotometric determination of desoxyribosenucleic acid in megaloblasts of pernicious anemia. Blood, 6: 344, 1951
130. Rogers, L L. and Shive, W Biological transformations as determined by competitive analogue—metabolite growth inhibitions VII. Relation of purines and thymine to folic acid J Biol Chem, 172: 751, 1948
131 Ross, G I M · Vitamin B_{12} assay in body fluids Nature, 166: 270, 1950.
132 Roughton, F J W. Some recent work on the chemistry of carbon dioxide transport by the blood Harvey Lectures, 39: 96, 1943–44.
133 Sabin, F. R., Miller, F R, Smithburn, K C., Thomas, R M and Hummell, L E Changes in the bone marrow and blood cells of developing rabbits J. Exp Med, 64: 97, 1936
134 Sabine, J C The cholinesterase of erythrocytes in anemia Blood, 6: 151, 1951
135 Sakami, W The conversion of formate and glycine to serine and glycogen in the intact rat J Biol Chem, 176: 995, 1948
136 Sakami, W and Welch, A D · Synthesis of labile methyl groups by the rat in vivo and in vitro J Biol Chem, 187: 379, 1950
137 Sauberlich, H. E. and Baumann, C A A factor required for the growth of leuconostoc citrovorum J Biol Chem, 176: 165, 1948
138 Sauberlich, H E The effect of folic acid upon the urinary excretion of the growth factor required by leuconostoc citrovorum J Biol Chem, 181: 467, 1949
139 Schleicher, E M The origin and nature of the Cabot ring bodies of erythrocytes J Lab & Clin Med, 27: 983, 1942
140 Schlenk, F Chemistry and enzymology of nucleic acids Adv in Enzymology, 9: 455 Interscience Publishers, Inc, New York, 1949
[illegible] of bone marrow and
[illegible] opterin
[illegible] 8, 1950
[illegible] evalua-
[illegible] Med,
35: 894, 1950
144 Schwartz, S O and Blumenthal, S A Exogenous hemachromatosis resulting from blood transfusions Blood, 3: 617, 1948
145 Shemin, D, London, I M and Rittenberg, D The in vitro synthesis of heme from glycine by the nucleated red blood cell J Biol Chem, 173: 799, 1948, ibid 183: 749, 757, 184: 745, 755, 1950

191. WRIGHT, L. D., MILLER, C. S., SKEGGS, H. R., HUFF, J. W., WEED, L. L. AND WILSON, D. W.: Biological precursors of the pyrimidines. J. Am. Chem. Soc., 73: 1898, 1951.

192. WYMAN, J., JR.: Relation of physiological function and molecular structure in hemoglobin. Fed. Proc., 7: 502, 1948.

193. YOUNG, L. E. AND LAWRENCE, J. S.: Maturation and destruction of transfused human reticulocytes. Evaluation of reticulocyte experiments for measurement of hemoglobin metabolism. J. Clin. Invest., 24: 554, 1945.

194. ZUELZER, W. W. AND OGDEN, F. N.: Folic acid therapy in the macrocytic anemias of infancy. Proc. Soc. Exp. Biol. & Med., 61: 176, 1946.

livery of new red cells to blood by high concentrations of inspired oxygen. J. Clin Invest , 28: 1544, 1949.

169. UNGLEY, C. C.: Thymidine and vitamin B_{12} in pernicious anemia Lancet, 1 164, 1949.

170. UNGLEY, C. C : Absorption of vitamin B_{12} in pernicious anemia II. Oral administration with normal gastric juice. Br. Med. J , 2: 908, 1950

171. UNGLEY, C C. AND THOMPSON, R. B.. Vitamin B_{12} and folic acid in megaloblastic anemias of pregnancy and the puerperium Br. Med. J , 1: 919, 1950

172. VALENTINE, W.: Quantitative biochemical studies on leucocytes in man, a review. Blood, 6: 845, 1951.

173. VALLEE, BERT AND ALTSCHULE, M. D : Editorial. trace metals in blood with particular reference to zinc and carbonic anhydrase Blood, 4: 398, 1949

174. VILTER, C. F., VILTER, R W. AND SPIES, T. D : The occurrence of combined system disease in persons with pernicious anemia during treatment with the Lactobacillus casei factor (Folic acid). J. Lab. & Clin. Med., 32: 335, 1947.

175 VILTER, R. W., WOOLFORD, R M. AND SPIES, T. D.: Severe scurvy, a clinical and hematologic study. J Lab. & Clin. Med., 31: 609, 1946.

176. VILTER, R. W.: Vitamin C and anemia Symposia on nutrition of the anemias Nutritional Anemias, Vol. I, A. Lejwa, Editor. Robert Gould Research Foundation, Inc , 1947.

177. VILTER, R W., HORRIGAN, D., MUELLER, J F , JARROLD, T , VILTER, C F., HAWKINS, V AND SEAMAN, A.· *Studies on the relationship of vitamin B_{12},* folic acid, thymine, uracil, and methyl group donors in persons with pernicious anemia and related macrocytic anemias Blood, 5: 695, 1950

178 VONBONSDORFF, B : Pernicious anemia caused by Diphyllobothrium latum, in the light of recent investigations Blood, 3: 91, 1948

179 WALDENSTRÖM, J · Successful treatment of liver refractory anemia with synthetic Lactobacillus casei factor Blood, 2: 426, 1947

180. WARREN, C. O : Respiration and glycolysis of rabbit bone marrow in serum in relation to cellular components Am J Physiol , 131: 176, 1940

181. WATSON, C J , GRINSTEIN, M AND HAWKINSON, V Studies of protoporphyrin IV A comparison of the erythrocyte protoporphyrin concentration with the reticulocyte percentage under experimental and clinical conditions J Clin Invest., 23: 69, 1944

182. WATSON, C J The erythrocyte coproporphyrin and its variation in respect to protoporphyrin and reticulocytes in certain of the anemias Trans Assoc Am Phys , 63: 219, 1950

183 WEBSTER, S H Heinz body phenomenon in erythrocytes, a review Blood, 4: 479, 1949

184 [illegible] macrocytic anemias [illegible]

185 [illegible] ience, 107: 398, 1948

186 WILKINSON, J F Folic acid Brit Med J , 1: 771, 822, 1948

187. WILLS, L · Pernicious anemia, nutritional macrocytic anemia and tropical sprue Blood, 3: 36, 1948

188. WINTROBE, M. M , CARTWRIGHT, G E , LAHEY, M E AND GUBLER, C J The role of copper in hemopoiesis Trans Assoc Am Phys , 64· 310, 1951

189. WINTROBE, M M Clinical Hematology Lea and Febiger, 1951

190. WOODRUFF, C. W Tyrosine metabolism in infantile scurvy J. Lab & Clin Med., 36: 640, 1950

PHARMACOLOGY

The available chemotherapeutic agents do not inhibit specifically the growth of cancer, but act on the normal functions or the special properties retained by the cancer cells from their tissue of origin. For this reason, the drugs used against cancer are pharmacologically active agents, which modify the growth of certain normal as well as cancer cells. The mechanisms whereby these agents act fall into four divisions (58).

1. The *general cell poisons* include *nitrogen mustard* (HN2), *triethylene melamine* (TEM), *urethane*, *potassium arsenite* and *radio-active phosphorus* (P^{32}). These agents inhibit growing tissues. The bone marrow and, in some animals, the intestinal mucosa, are particularly susceptible to their action. Excessive doses will cause a severe depression in hematopoietic function.

2. The *antimetabolites* include the *folic acid antagonists*. The antimetabolites are similar in structure to certain normal precursors necessary for cell growth, and an excess of the abnormal compound will interfere with the proper utilization of the normal precursor, and thus induce cellular injury. The tissues most susceptible to the folic acid antagonists are the bone marrow and the lining of the digestive tract.

3. *Hormones* are essential to maintain the growth and functional activity of certain tissues, and induced hormonal imbalances may result in regressions in certain types of tumors. The hormones in common use are the *androgens*, *estrogens*, *adrenocortical hormones* (*cortisone*) and *corticotropin* (ACTH)

4. *Specific cell poisons* injure certain cancer cells because of the special ability of these cells to concentrate certain substances The illustrative agent in this group is radioactive iodine (I^{131}), which concentrates in functionally active thyroid cancers and then destroys the cells by radiation

General Cell Poisons

NITROGEN MUSTARD AND ALLIED SUBSTANCES

Methyl-bis-(B-chloroethyl)-amine hydrochloride, known by the code name HN2, has been in general clinical use since 1946 (91), and its pharmacological properties are well-established (84) The formula of HN2, and its transformation in aqueous solution to a reactive imonium compound is shown in figure 1 HN2 has specific and injurious effects on cells, as evidenced by its interference with nucleic acid formation and cell division, and by its mutagenic and carcinogenic activity In mammals, at LD_{50} doses, it produces bone marrow aplasia with leukopenia, decrease in platelet count, anemia, destruction of the lymphatic tissue and ulceration of the intestinal mucosa associated with severe diarrhea Death usually occurs within 4 to 7 days. HN2 has been shown to act directly on the tissues

Chemical Agents Used in the Treatment of Inoperable and Far-Advanced Neoplastic Disease*

DAVID A. KARNOFSKY, M.D.

INTRODUCTION

As a result of pains-taking investigations, several chemical agents have been found which will temporarily restore normal activity, relieve discomfort and prolong life in patients with certain types of inoperable or far-advanced cancers (31, 42, 53, 56, 58, 90, 102, 110). The physician, along with his knowledge of antibiotics, hypnotics, cardiac glucosides, diuretics, antispasmodics, vitamins, etc, now must become familiar with a new branch of pharmacology, the *general and specific cell poisons,* drugs capable of selectively inhibiting the growth of certain tissues The growth-inhibiting drugs have undergone intensive pharmacological study, and an understanding of their pharmacological properties and therapeutic indications makes their administration to man no more hazardous than that of many other substances now in routine clinical use. The physician interested in the management of inoperable cancer should be able to apply these agents of proved value in an appropriate and effective manner, and with a proper awareness of their limitations.

The attempt to treat cancer with systemically administered drugs has a long and undistinguished history (46, 123), the effort having been associated with empiricists and charlatans, active proponents and dispensers of secret and irrational therapeutic agents While physicians seem to retain their critical faculties in other fields of medicine, in treating cancer some, along with the public, are willing to entertain and even support therapeutic claims of the most fantastic nature This report will present those chemotherapeutic agents which have been shown to be of value in cancer, these drugs do not cure cancer and they are not applicable universally, but they have survived intensive clinical trials and their range of effectiveness is supported by evidence from numerous independent clinics Their beneficial influences in patients are not, in an absolute sense, *impressive, but* they mark the initial contributions of a concerted effort to find effective chemicals for the treatment of cancer.

* [illegible] Institute of the

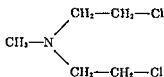
HN2 is injected usually into the tubing of a saline infusion, in order to be certain that all the drug enters the vein; and this procedure apparently reduces the incidence of venous thrombosis at the site of injection. If the solution of HN2 extravasates it produces a severe local reaction, and the involved area becomes red, swollen and then indurated; local discomfort may persist for 4 to 6 weeks or longer. The injection is best given in late afternoon or early evening, so that the patient may sleep during the major period of nausea, and intramuscular barbiturates appear to be the most satisfactory method of allaying the vomiting. Occasionally patients feel that pyridoxine or dramamine reduces the gastrointestinal distress. The vomiting induced by HN2 has had no serious consequences, except in patients with bleeding tendencies

The important toxic effect of HN2 is depression of the bone marrow. At therapeutic doses, a moderate leukopenia and thrombocytopenia occur; the maximal fall occurring one to two weeks after HN2. Following this low-point, recovery is rapid, and within 4 to 6 weeks the blood cells have returned to normal. With excessive dosage, a severe depression of the bone marrow occurs, with a syndrome of leukopenia, thrombocytopenia, bleeding, anemia and fever, and this may terminate in death, usually 2 to 3 weeks after the course of treatment If the patient survives this period, however, recovery may be rapid. In patients with bone marrow depression or infiltration due to disease, HN2 sometimes may induce further damage, and recovery may be very slow or absent The protracted bone marrow depression following HN2 is not primarily due to the drug, but to the impaired regenerative capacity of the bone marrow following injury Five degrees of hematopoietic injury have been seen following treatment with nitrogen mustard or allied substances (57):

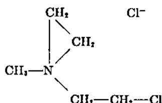
1) Moderate leukopenia developing in one to two weeks and followed by rapid recovery
2) Moderate to severe, but transient, leukopenia and a temporary fall in hemoglobin levels These values return to their original levels within two to four weeks This is associated with a fall in thrombocyte count, but there is no evidence of bleeding
3) Severe leukopenia and thrombocytopenia with bleeding and a fall in hemoglobin level which is aggravated by the bleeding tendency Evidences of hemorrhage consist of generalized petechiae, ecchymoses, particularly following slight injury, and nasal, oral, rectal and renal bleeding The bone marrow depression is transient, and evidences of active bone marrow regeneration appear in two to six weeks
4) Severe and prolonged bone marrow depression, with persistent leukopenia, anemia, thrombocytopenia and bleeding. Patients may continue in this state for several months after treatment These prolonged bone marrow depressions seem to occur in patients with

damaged, although some of its injurious effects on lymphatic tissue may be secondary to stimulation of the adrenal cortex. Following intravenous injection, HN2 is rapidly removed from the blood stream, and it is presumably fixed in the tissues within 5 minutes HN2 is injurious to both normal and neoplastic cells, and in many respects its effects parallel those of x-rays There is considerable evidence, however, that, at comparable degrees of tissue damage induced by HN2 and x-rays in mammals, the tissues recover more rapidly from the injury produced by nitrogen mustard; therefore, it is safer to administer the maximal tolerated dose of HN2 as compared to total body irradiation (56).

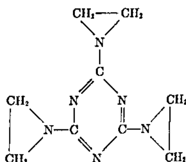
HN2 is a fine white powder which is diluted with distilled water just



Methyl-*bis*-(2-chloroethyl)-amine (HN2)



Ethylenimonium transformation of HN2



2,4,6-triethylenimino-*s* triazine (triethylene melanine) (TEM)

FIG. 1. COMPARISON OF THE STRUCTURAL FORMULAS OF METHYL-BIS-(2-CHLOROETHYL)-AMINE AND TRIETHYLENE MELAMINE

before use. The usual dose in a course of treatment in man is 0 4 mg per kg of body weight (25 to 30 mg in the average adult) given as a single dose, or more often divided into 2 to 4 daily doses Some patients with evidence of impaired hematopoiesis may safely tolerate only 0 2 mg per kg in a course of treatment, whereas patients with normal or hyperactive bone marrow may be given as much as 0 8 mg per kg. The degree of bone marrow depression induced by HN2 is the limiting factor in treatment In many instances it is desirable to give HN2 to the point of maximal tolerance in the hope of producing clinical improvement; the dosage is determined by repeated study of the formed elements of the blood during the course of treatment, and the amount of HN2 given must be worked out for each patient. Frequently, each injection of HN2 will cause nausea and vomiting, usually one to 8 hours after the injection, nausea and vomiting occur sooner with large single doses, but their severity is not increased

the systemic toxicity of HN2. The efficacy of this procedure has not been proved.

Triethylene melamine (TEM) (structural formula shown in fig. 1) is a compound closely allied pharmacologically to the nitrogen mustards because of the reactive ethyleneimine groups TEM differs from HN2 in its effect on animals chiefly in that it lacks the convulsant and cholinergic activity of HN2 (86). Its administration in man by either the intravenous or oral route causes very little nausea or vomiting. TEM is about 2 to 3 times as active as HN2 by weight, and the total course of TEM by the intravenous route ranges from 8 to 12 mg (0.12 to 0 2 mg. per kg of body weight (57, 86)). The usual course of TEM is 0 04 mg. per kg. on three to four successive days. A single 0 04 mg per kg dose of TEM usually does not induce nausea or vomiting, but larger single doses have occasionally caused severe vomiting. TEM is a white, water-soluble powder, which is made up freshly in physiological saline just before use It may contain insoluble polymer which causes an opalescent solution. The injection may be given in a 0 5 mg per cc. solution by direct intravenous injection, since TEM rarely causes venous thrombosis The extravasated solution produces local tissue damage similar to that caused by HN2 TEM also causes bone marrow depression, in a pattern paralleling that of nitrogen mustard Its use entails all the hazards, and requires all the precautions already described for HN2

TEM is prepared in scored tablets containing 5 mg. with excepient for oral use Since TEM is very reactive with organic substances, it is taken early in the morning, one hour before breakfast, with plain water The single daily dose is usually 2.5 to 5 mg , and about 50 per cent of the patients can tolerate this dose without difficulty, the remainder at some time following a dose may complain of nausea, but vomiting is infrequent The effective total dose of oral TEM is variable, ranging from the extremes of 10 to 150 mg , although the average patient usually tolerates 20 to 40 mg during a course of treatment Oral TEM consequently must be administered slowly and with care, and 3 to 4 weeks are often necessary to deliver an adequate course of treatment In table 1 a dosage schedule for oral TEM is presented The maximal bone marrow depression may follow the last dose of TEM by 2 to 4 weeks, and blood counts must be taken each week, and further dosage ordered on the basis of the white cell count and the clinical situation By means of protracted treatment with oral TEM, the maximal tolerated dose, based on bone marrow depression, may be given much more precisely than with intravenous HN2 Furthermore, a patient may be given further TEM at the earliest signs of relapse, and be maintained on this treatment as long as the patient continues to respond The bone marrow depression following oral TEM is similar to that seen after intravenous TEM or

far-advanced disease and previous evidence of hematological abnormality.

5) Bone marrow depression with severe bleeding and fever, resulting in death within two to three weeks after treatment, presumed to be the direct result of overdosage with HN2

There are no known specific measures for reversing the bone marrow depression induced by HN2. Blood transfusions are given for the bleeding and anemia, and antibiotics are used as prophylaxis against infection. Smith et al. (106) have observed prolongation of clotting time, presumably due to hyperheparinemia, in HN2 toxicity, and toluidine blue and protamine have been reported to control the bleeding tendency.

Complaints which have occurred sometimes during HN2 therapy are weakness, anorexia, drowsiness, a metallic taste in the mouth and diarrhea. Zanes et al. (124) have described a maculopapular pruritic skin eruption following HN2.

The use of nitrogen mustard is considered frequently in conjunction with local x-ray therapy, and some general observations may be made on these two forms of treatment During the initiation of x-ray therapy, local edema may occur, and this can be dangerous if the tumor is growing in certain vulnerable areas, such as in the thorax. Edema in the tumor-bearing area, in our experience, has not occurred following HN2 and, in some patients, a course of HN2 prior to x-ray therapy is indicated Furthermore, in mortality studies on mice, HN2 given prior to irradiation produced an additive lethal effect, whereas in the reverse sequence, the total effect was less than additive (59) Webber et al. (121), however, have reported more recently, on the basis of cytological changes in the rat intestine, that HN2 is more cytotoxic when given 24 hours after local intestinal irradiation The sequential use of nitrogen mustard and x-ray therapy obviously requires further study The application of x-ray therapy to limited, localized areas of neoplastic disease has not aggravated the bone marrow depression produced by HN2, in fact, in many cases treated by HN2 followed by x-ray therapy, leukopenia due to HN2 developed and then improved while x-ray therapy was being given

It has been shown in animals that the bone marrow is protected from the cytotoxic effect of HN2 by depriving the bone marrow of its blood supply during and for a few minutes following the intravenous injection of HN2 (61) On this basis, in selected cases, sphygmomanometer cuffs inflated above arterial pressure have been applied to both thighs and one arm during and for 5 minutes following the rapid intravenous injection of HN2 It is hoped that this measure may protect some of the peripheral marrow which, unfortunately, is normally hypoactive, and thus diminish

ment ranges from 20 to 200 grams or more. Excessive amounts will cause severe bone marrow depression with leukopenia, thrombocytopenia, bleeding and anemia, which may lead to death. Also, several instances of fatal

TABLE 1

Dosage Schedules in Initial Course of Oral Therapy with Triethylene Melamine (57)

	FIRST DOSE (1ST WEEK)	SUBSEQUENT TREATMENT* (2ND TO 4TH WEEKS)
	mg	
Usual schedule	10†	Each week a decision is made to give 5 or 10 mg or temporarily to stop therapy; at the end of the 4th week, each patient has usually received a total of 20 to 40 mg
Exceptions 1 Poor general condition, particularly with bone marrow depression 2 Lymphosarcoma 3 Chronic lymphatic leukemia	5	Further treatment is given cautiously, these patients may tolerate only 5 mg or as high as 40 mg over 4 weeks, excessive dosage is to be avoided in susceptible patients
Carcinoma with apparently intact bone marrow	15	Treatment may sometimes be given more rapidly, after a preliminary trial of 2 weeks, if no effects on blood count or clinical course occur, larger doses are given, at the end of the 4th week, the total dose may range from 30 to 150 mg

* In most instances, clinical improvement, if it is to occur, appears within four weeks after starting triethylene melamine therapy If improvement has occurred, further treatment depends on the duration of the patient's response and his tolerance to the drug If no improvement has occurred and there is evidence of severe bone marrow depression, further triethylene melamine therapy will probably not be effective, however, if the white blood cell count and hemoglobin level are unchanged after four weeks, larger doses of triethylene melamine are given

† The usual single daily dose is 5 mg , but if patient complains of nausea or vomiting it is reduced to 2 5 mg

hepatitis have been associated with urethane therapy (80). The patient should be followed carefully while on urethane therapy, and blood counts taken at least at weekly intervals for early evidence of bone marrow depression An adequate trial of therapy may require cautious but definite and sometimes repeated, depressions of the white cell count in certain conditions Thus, despite its relatively low toxicity, urethane must be administered as carefully as some of the more active cytotoxic agents

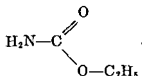
HN2. Oral TEM is an effective and convenient substitute for nitrogen mustard therapy in the ambulatory patient.

OTHER NITROGEN MUSTARDS AND ALLIED SUBSTANCES

A number of other agents has been tried clinically, but they have not shown any therapeutic or practical advantage over HN2 or TEM; some of the trials, however, are not yet completed. These chemicals are summarized in table 2.

Urethane

Urethane or ethyl carbamate is a general cell poison with the structure



It has been used clinically as a weak hypnotic for many years, but in 1946 Haddow and Sexton (47) *reported that urethane would inhibit the* growth of mouse and rat tumors On the basis of this observation Paterson et al. (82) administered urethane to patients with various forms of neoplastic disease, and observed clinical and hematological improvement in chronic myelogenous and lymphatic leukemia These observations prompted more detailed studies on the cytotoxic activity of urethane in animals. While it has a low toxicity by weight, at adequate doses and prolonged administration, urethane will produce bone marrow depression (21, 76), reduce mitotic activity and cause cellular degeneration of the intestinal epithelium (30), induce lung tumors in mice (79), and prolong the life of animals with transplanted (11) and spontaneous leukemia (69). The urethane molecule has a high biological specificity, and closely related chemical structures are inactive (11, 105) There is, as yet, no satisfactory explanation of the mechanism of action of urethane, but it presumably interferes with nucleoprotein synthesis, acting as an antimetabolite or an enzyme inhibitor in inhibiting the growth of rapidly dividing cells

Urethane has been given by the oral, intramuscular and intravenous routes in man. The oral route is preferred, and urethane may be given in enteric-coated tablets or as a 25 per cent solution, sometimes *added to a* palatable drink. The usual daily dose ranges from 2 to 4 grams per day, in 3 to 4 divided doses, and an adequate clinical trial may require one month

symptoms, but occasionally they are so severe as to necessitate the cessa- of urethane therapy. The total dose of urethane during a course of treat-

(8)	1 3 propane diamino-2-chloro NNN′N′ tetrakis (2-chloro-ethyl) 2HCl (SK 137)	$N(CH_2CH_2Cl)_2$–CH_2–$CHCl$–CH_2–$N(CH_2CH_2Cl)_2$	3–4	200–300	—	Occasional toxic psychosis *Not in use*
(74)	B-naphthyldi-2-chloroethylamine (R48)	naphthyl–$N(CH_2CH_2Cl)_2$	—	—	300–400 mg /day × 10 to 20 days	Dosage control difficult Some nausea and vomiting. *Not in general use*
(112)	3-bis(B-chloroethyl)aminomethyl-4-methoxy-methyl-5-hydroxy-6-methylpyridine 2HCl (SK 1424)	OH, CH_2OCH_3, $CH_2N(CH_2CH_2Cl)_2$, CH_3, N (pyridine ring); 2 HCl	5	250–400	200 mg /day × 10 to 20 days	Dizziness, severe nausea and vomiting when taken by mouth *Not in use*

TABLE 2

Compounds, Related to Nitrogen Mustard, Which Have Been Used in Man

REF *	NAME	FORMULA	APPROXIMATE DOSE PER COURSE OF TREATMENT			CLINICAL STATUS OF DRUG
			Intravenous		Oral (mg /total)	
			Mg /kg	Mg /total		
(45)	tris-B chloroethyl amine (HN3)	$N(CH_2CH_2Cl)_3$	0 2–0 3	15–25	—	Activity similar to HN_2, *not in use*
(8)	1·3 propane diamine NNN'N'tetrakis-(2-chloroethyl)·2HCl (Sk 136)	$(ClCH_2CH_2)_2N{-}CH_2{-}CH_2{-}CH_2{-}N(CH_2CH_2Cl)_2 \cdot 2HCl$	0 4–0.6	25–40	—	Dizziness, some nausea and vomiting *Not in general use*

(8)	1·3 propane diamino-2-chloro NNN′N′ tetrakis (2 chloroethyl) 2HCl (SK 137)	$(ClCH_2CH_2)_2N-CH_2-CHCl-CH_2-N(CH_2CH_2Cl)_2$	3–4	200–300	—	Occasional toxic psychosis *Not in use*
(74)	B-naphthyldi-2-chloroethylamine (R48)	$C_{10}H_7-N(CH_2CH_2Cl)_2$	—	—	300–400 mg /day × 10 to 20 days	Dosage control difficult. Some nausea and vomiting. *Not in general use*
(112)	3-bis(B-chloroethyl)aminomethyl-4-methoxy-methyl-5-hydroxy-6-methylpyridine 2HCl (SK 1424)	pyridine ring with OH, CH_2OCH_3, $CH_2N(CH_2CH_2Cl)_2$, CH_3, N; 2 HCl	5	250–400	200 mg /day × 10 to 20 days	Dizziness, severe nausea and vomiting when taken by mouth *Not in use*

TABLE 2

Compounds, Related to Nitrogen Mustard, Which Have Been Used in Man

REF *	NAME	FORMULA	APPROXIMATE DOSE PER COURSE OF TREATMENT			CLINICAL STATUS OF DRUG
			Intravenous		Oral (mg /total)	
			Mg /kg	Mg /total		
(45)	tris-B chloroethyl amine (HN3)	CH_2CH_2Cl $N-CH_2CH_2Cl$ CH_2CH_2Cl	0 2–0.3	15–25	—	Activity similar to HN_2, *not in use*
(8)	1·3 propane diamine NNN'N'tetrakis-(2-chloroethyl)·2HCl (Sk 136)	CH_2CH_2Cl N CH_2CH_2Cl CH_2 CH_2 CH_2 CH_2CH_2Cl N CH_2CH_2Cl 2HCl	0.4–0 6	25–40	—	Dizziness, some nausea and vomiting. *Not in general use*

Potassium Arsenite ($KAsO_2$)

Fowler's solution (1 per cent As_2O_3) has been used widely in the treatment of chronic myelogenous and lymphatic leukemia The dosage is 3 drops three times daily, and each dose is increased at the rate of one drop daily until clinical improvement occurs or signs of toxicity, such as nausea and vomiting, diarrhea, or skin eruptions develop When this point, or when a maximal dose of 20 drops three times daily, is reached, the dose is gradually decreased, and a maintenance dose is established of about 5 drops three times daily (36)

Antimetabolites

FOLIC ACID ANTAGONISTS

The structures of folic acid and one of its 4-amino derivatives (4-amino pteroylglutamic acid) are shown in figure 2. The replacement in the folic acid molecule of the hydroxyl group in the 4 position by an amino group results in a chemical of extraordinary toxicity which, on administration to animals, will produce the pathological effects of an acute folic acid deficiency Animals develop bone marrow depression with megaloblastosis, associated with leukopenia, thrombocytopenia and anemia in the peripheral blood, and ulcerations appear in the gastro-intestinal tract (85) This effect on rapidly growing tissues is also reflected in the inhibition in growth of experimental animal tumors and mouse leukemia (111). The mechanism of action of 4-amino PGA has been reviewed most recently by Welch and Heinle (122). Enormous doses of folic acid will provide slight protection only against minimal toxic doses of 4-amino PGA On the other hand, folinic acid (citrovorum factor), which has been synthesized as the structure shown in figure 2, will protect competitively, but principally non-competitively, against very large doses of 4-amino PGA This would indicate that the 4-amino derivatives of folic acid act by blocking the conversion of folic acid to folinic acid and, probably, but to a lesser extent, by direct competition with folinic acid It is well-established that folinic acid is necessary to formate transfer, a step in the synthesis of purines and pyrimidines By a block in this process, synthetic processes related to nucleic acid formation are inhibited, and cell growth ceases Diversified growing systems, such as bacteria (1), drosophila (43), and the chick embryo (62) have been shown to be susceptible to folic acid deficiency and the folic acid antagonists In animals, folinic acid will protect against 4-amino PGA if the injections are given close together, if the intervals are too far apart folinic acid will not reverse the cell damage already initiated by 4-amino PGA If, however, evidences of moderate 4-amino PGA toxicity are present, the continuation of 4-amino PGA with the addition of folinic acid will inhibit the progressive manifestations of 4-amino PGA toxicity.

TABLE 2—*Continued*

REF*	NAME	FORMULA	APPROXIMATE DOSE PER COURSE OF TREATMENT			CLINICAL STATUS OF DRUG
			Intravenous		Oral (mg /total)	
			mg /kg.	mg /total		
(7)	Triethylene phosphoramide (TEPA)	$(CH_2CH_2N)_3P{=}O$	1 5–2.5 (IM)	80–120 (IM)	—	Under study
(119)	1,4-dimethane sulphonyl oxybutane (G T 41)	$CH_3SO_2O{-}CH_2{-}CH_2{-}CH_2{-}CH_2{-}OSO_2CH_3$	—	—	150–250 mg total	Under study

but there has been no demonstrated qualitative change in its biological activity in mammals at effective doses. Some of the derivatives of 4-amino PGA, and their comparable dosages in man are given in table 3.

The folic acid antagonist most commonly used in our clinic is 4-amino N^{10}-methyl PGA (A-methopterin). It is available in 2.5 mg tablets for oral use. The dosage in children ranges from 1.25 to 5.0 mg per day, usually given as a single dose. Adults may tolerate 5 to 10 mg daily. There is considerable variation among patients in the dose tolerated; the reason for these individual differences is not known, but there is evidence that A-methopterin is more toxic in patients with impaired renal function, presum-

TABLE 3

Derivatives of 4-Amino PGA Studied for Clinical Activity (10)

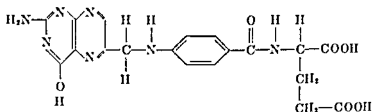
COMPOUND		DOSAGE	
Chemical name	Trade name	Children	Adults
		mg*	mg*
4-Amino-PGA	Aminopterin	0.5–1.0	1.0–3.0
4-Amino-N^{10}-methyl-PGA	Amethopterin	2.5–5.0	5.0–10.0
4-Amino-9 methyl-PGA	Aninopterin	2.5–5.0	5.0
4-Amino-9,10 dimethyl-PGA	Adenopterin	1.25–2.5	2.5–5.0
4-Aminopteroylaspartic acid	Aminoanfol	20–50	50–75
4-Aminopteroyltriglutamic acid	Aminoteropterin	0.25–0.5	0.5–1.0

* In our experience, aminopterin and amethopterin are effective in these doses whether given orally or by subcutaneous injection. The dosages of the other derivatives of PGA are given for subcutaneous injection. This does not imply, however, that they are ineffective by mouth, but merely that they were given parenterally in our series.

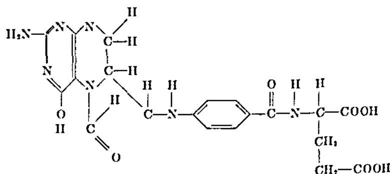
ably due to the more prolonged retention of the agent. Usually A-methopterin in children is started at 2.5 mg per day, and the patient watched closely for signs of toxicity and clinical improvement. Depending on the urgency of the situation and the patient's response, dosage may be continued at this level, decreased or increased. Usually the dosage is increased after a 10 day trial at the starting level, unless signs of toxicity intervene. The principle of therapy is to induce a clinical remission or continue the *treatment at increasing doses until evidences of toxicity appear. Treatment* is stopped at this point, but may be restarted as soon as the signs of toxicity clear. The early manifestations of 4-amino PGA toxicity are extremely important in guiding therapy, and they should be thoroughly understood. In their usual order of frequency of occurrence, they are:

Mouth Lesions. These are usually the earliest signs of toxicity, and appear as whitish areas of necrosis on the lips, gingivae, palate or uvula. They

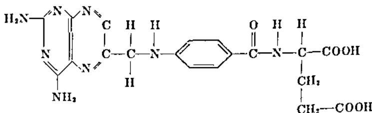
The 4-amino group is essential for the anti-metabolite activity of 4-amino PGA, but a number of active analogues with substitutions in the 9 and 10 position and in the glutamic acid side-chain have been prepared. The modifications have decreased, to various degrees, the activity of the antagonist,



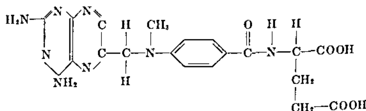
Folic acid



Folinic acid (synthetic citrovorum factor)



4-Amino PGA (Aminopterin)



4-Amino N^{10} methyl PGA (A-methopterin)

FIG 2. FORMULAE OF FOLIC ACID, FOLINIC ACID, 4-AMINO PGA AND 4-AMINO N^{10} METHYL PGA

Sex Hormones

The male and female sex hormones are used widely in the treatment of cancer. These agents are available in several different forms, but they appear to have approximately the same therapeutic activity at the proper dosage.

TABLE 4

Dosages of Steroid Hormones and Corticotrophin in Cancer Therapy

	ROUTE	USUAL DOSE	
		Breast cancer	Prostate cancer
Estrogens			
Diethylstilbestrol	oral	5 mg t i d.	1–5 mg t i d.
Ethinyl estradiol (Estinyl)	oral	1 mg t i d.	0 1–0 5 mg t i d
Sodium estrone sulfate	oral	10 mg t i d.	—
Estradiol benzoate	i m.	5 mg t i w	—
Androgens			
Testosterone propionate	i.m	50–100 mg t.i.w.	
Methyl testosterone	oral	50–200 mg. daily	
Methyl androstenediol	oral	200 mg daily	
Dihydrotestosterone	i m.	100 mg 6× weekly	
		Adults	Children
Adrenocortical and Pituitary Hormones			
Cortisone	oral	200 mg /day (in divided doses)	150–200 mg /day (in divided doses)
ACTH	i m	200 mg /day (in divided doses)	100 mg /day (in divided doses)
"	i v	50 mg /day (continuous infusion)	25 mg /day (continuous infusion)

ANDROGENS

The principal androgen in use is testosterone propionate (table 4). The usual recommended weekly dose in the treatment of breast cancer ranges from 150 to 300 mg intramuscularly, given as 50 to 100 mg. doses three times weekly. The lower dosage schedule is probably as effective as the higher one (17). Methyl testosterone by mouth is administered at somewhat higher doses, methylandrostenediol recently has been reported as having the therapeutic activity of testosterone without as marked virilizing effects (63) but this conclusion is not supported by Nathanson and Kelley (77) and Segaloff et al (100) Dihydrotestosterone is now under study as pos-

may be preceded by a reddened area, and the lesions are painful. Treatment should be stopped when mouth lesions appear, and they usually heal in 3 to 4 days. Occasionally the mouth lesions may appear coincidentally and are not due to treatment, and if this is strongly suspected and further treatment essential, A-methopterin has been continued.

Ulcerations of the Digestive Tract. Ulcerations may also occur in the esophagus, small intestine and colon, presumably related to the ulcerations of the oral mucosa. Treatment should be stopped temporarily if the patient complains of abdominal cramps or diarrhea. If further A-methopterin is given bloody diarrhea may develop

Bone Marrow Depression. The antifolics can induce severe depression of all the bone marrow elements, with megaloblastosis (118), and this is reflected in the peripheral blood as leukopenia, thrombocytopenia, hemorrhagic tendencies and anemia. The white cell count and hemoglobin should be followed closely during treatment, and treatment temporarily stopped if the white cell count or hemoglobin level show an unexpected decline.

Miscellaneous or inconstant effects include: a *maculoerythematous sensitivity reaction of the skin,* occurring chiefly on the trunk (this is usually relieved by antihistaminics, but treatment should be discontinued temporarily), *hyperpigmentation, interruption in hair growth and alopecia,* and evidences of *increased susceptibility to infection*

OTHER METABOLIC ANTAGONISTS

Several other metabolic antagonists are being studied clinically because of their inhibitory effects on the growth of transplanted tumors and leukemia in mice, these agents have shown some evidence of therapeutic activity, but their clinical value has not been established.

2,6-Diaminopurine is presumably an antagonist of adenine although, more probably, it interferes with cell growth by producing an imbalance among the precursors used in nucleic acid synthesis In man, doses of 5 to 20 mg. per kg by mouth continued over several days to weeks can induce bone marrow depression and, occasionally, a therapeutic effect in leukemia (10). Unfortunately, it also causes severe nausea and vomiting which have precluded an extensive clinical trial

Diamino dichlorophenyl pyrimidine has pharmacological properties similar to those of the folic acid antagonists, and it has inhibited the growth of transplanted mouse tumors (9, 15) In preliminary clinical studies it appears to be somewhat similar to the 4-amino PGA derivatives in its therapeutic effects, but its toxic manifestations have been less consistent and predictable than those occurring with the 4-amino PGA derivatives, dosage control is, therefore, more difficult

situation has been reviewed carefully, and the patient should then be followed closely.

The actions of the estrogenic hormones on breast cancer are unexplained; they presumably produce some type of hormonal imbalance or they may inhibit the anterior pituitary. The growth of some prostatic carcinomas may be dependent on androgenic stimulation, and estrogen therapy may counteract androgenic activity directly and/or inhibit the anterior pituitary so that androgen production is decreased.

ADRENOCORTICAL HORMONES

The adrenocorticotropic hormone of the pituitary gland (ACTH) stimulates the adrenal cortex to produce the adrenocortical steroids, and 17-hydroxycorticosterone (Kendall's Compound F) appears to be the principal steroid secreted. If large doses of exogenous ACTH are given, the adrenal cortex is greatly stimulated and the syndrome of hyperadrenocorticism results Cortisone acetate (11-dehydro-17-hydroxycorticosterone) is closely related to Compound F, and it produces systemic effects similar to those which follow ACTH administration.

The dosages used in the treatment of neoplastic disease are usually large (table 4) (83) Cortisone is given orally, beginning at a total dose of 150 to 200 mg per day in divided doses. If a therapeutic response occurs, or if undesirable physiological effects of cortisone appear, the dose is decreased and, if neither of these events occurs, the dose may be increased. In patients with extensive lymphomatous disease, the initial dosage should be smaller, since a rare patient may show rapid resolution of the neoplastic cells, and hyperurecemia and uremia, due to renal injury, may ensue. Equivalent doses of ACTH are in the range of 100 to 200 mg. daily, given intramuscularly in 4 to 8 divided doses. When a rapid response is desired, a continuous intravenous infusion of ACTH has been most effective, the ACTH is dissolved in 500 to 1000 cc of glucose in water and is dripped in slowly during the entire 24 hour period A dose of 25 mg in the child and 50 mg. in the adult each 24 hours appears to give a maximal response

During treatment the patient is followed carefully for evidences of fluid retention, electrolyte disturbance, hyperglycemia, elevation in blood pressure, and mental changes. The eosinophil count is checked as evidence of increased adrenocortical activity Patients are maintained on a low salt diet, and in adults testosterone propionate, 25 mg daily, has been given occasionally to counteract the catabolic effects of cortisone. In children excessive adrenocortical activity induced marked systemic changes; the child may acquire an increased appetite, gain weight rapidly and develop moon-facies, hirsutism and acne. In many instances the child's appearance is transformed in a gross and unattractive manner. Hypertension, transient

sibly having some advantage over testosterone. Nathanson and Kelley, however, state, "our studies thus far indicate that the best responses of breast cancer are intimately associated with the androgenic activity of a compound, irrespective of purely metabolic capabilities"

Testosterone does not ordinarily induce any reaction at the site of injection. It produces definite evidences of virilization, as shown by an increase in hirsutism, deepening of voice, acne, clitoral hypertrophy, vulvar itching, increased libido and temporary amenorrhea. Fluid retention may occur, and this may be a precipitating factor in *heart failure.* If there is evidence of marked fluid retention or heart failure, treatment should be interrupted temporarily, and the patient given a low sodium diet, with diuretics and digitalization if necessary. In patients with osseous lesions, androgen therapy may induce hypercalcemia. The first signs may be drowsiness, mental slowing and weakness and, if the condition is not recognized and androgen therapy is continued, it may be fatal; androgens should be stopped, calcium intake decreased and fluids forced in order to decrease the serum calcium level. It seems probably that most cases of hypercalcemia develop in patients with previous renal injury, but this is not an essential factor. An interesting response in some patients on androgen therapy is the increase in hemoglobin levels and red cell count (117). If the patient does *not develop serious untoward effects, treatment is continued for a minimal* period of 3 months

Nathanson and Kelley have reviewed the mechanism of action of androgens in breast cancer; while there are many lines of evidence supporting different theories, the correct answer is not clear. Androgens may act by interfering with the possible stimulating effect of estrogens on the cancer cells directly, or by inhibiting the formation of gonadotropic and, possibly, adrenocorticotropic hormones of the pituitary gland Furthermore, because of their anabolic effects, the androgens produce a general supportive action

ESTROGENS

Several preparations with estrogenic activity are available, of which diethylstilbestrol and ethinyl estradiol are most commonly used (table 4)

The side-effects associated with estrogen therapy are nausea, anorexia and occasionally vomiting, which usually subside after several weeks, enlargement of the breasts with tenderness, occasional uterine bleeding, which may be controlled by increasing the dose of estrogen, diarrhea, and incontinence, usually associated with stress In the male, impotence, shrinkage of the penis, and weight gain may occur Estrogens, as well as androgens, cause fluid retention, and this may precipitate heart failure in elderly patients, and hypercalcemia may develop in the presence of osseous lesions For these reasons, estrogen therapy should not be started until the clinical

P^{32} therapy, as with total body irradiation and nitrogen mustard administration, is damage to normal hematopoietic function, and, at excessive dosage, leukopenia, thrombocytopenia, bleeding and death can ensue Osgood (81) has described a method of maintenance therapy in leukemia, giving an initial dose of 20 and 40 microcuries per kg intravenously in chronic lymphatic and myelogenous leukemia, respectively, and giving further maintenance doses at stated intervals based on the condition of the patient

P^{32} therapy is a form of total body irradiation, although there is evidence of some degree of selective localization of P^{32} in growing cells and tissues and in bone.

Radioactive Iodine

I^{131} has a half-life of 8 days and it produces both beta and gamma radiation. It is the former which is chiefly destructive to tissue. I^{131} is used chiefly in the treatment of thyroid cancer. It localizes, as does normal iodine, in functioning thyroid tissue, and then its radiations damage the cells The patient must be studied and prepared carefully before I^{131} therapy can be considered by 1) establishing the diagnosis and extent of the disease, 2) determining the uptake of I^{131} by the tumor, 3) removing the normal thyroid tissue by surgery or by a thyroidectomizing dose of I^{131} in order to increase the uptake of I^{131} by the tumor cells and, 4) by the use of other adjuvants, such as thiouracil, to increase the avidity of the thyroid cancer for iodine. When the patient is ready, the usual dose of I^{131} is 100 to 200 millicuries by mouth although, in some cases, much higher doses have been used. The dose varies with the estimated amount of thyroid tissue present and its uptake of I^{131}, as measured by tracer doses

The chief hazards of treatment are 1) excessively rapid breakdown of neoplastic thyroid tissue with a "thyroid storm", 2) depression of the bone marrow function, although I^{131} is concentrated very effectively in the thyroid tissue, there may still be enough radioactivity in the blood to produce bone marrow injury and, 3) renal and gonadal injury due to radiation.

CLINICAL APPLICATIONS

The proper use of chemotherapeutic agents in the treatment of cancer requires an understanding of the various clinical types and manifestations of the disease, and the indications for and the results to be expected from surgery, radiation and chemotherapy Surgery and radiation therapy, thus far, offer the only possibilities for cure, and they should be applied, where feasible, as early and as effectively as possible There remains the large group of patients with systemic forms of neoplastic disease, or with recurrent or inoperable cancer, who require continuous care during the course

episodes of coma and mental disturbances have appeared in some cases. When a marked Cushing's syndrome develops, dosage is decreased or the agent stopped.

Adults show a lesser degree of moon-facies, acne and hirsutism, but fluid retention may be marked and have dangerous consequences Furthermore, cortisone appears to decrease resistance to infections, of both acute and chronic types, and these may complicate an otherwise favorable response. Cortisone may be of supportive value in relieving pain and producing a feeling of well-being, without having any demonstrable effect on tumor growth.

The adrenocortical hormones appear to be effective inhibitors of the growth of specific tissues, which may possibly be independent of their effects on electrolytes. Ragan et al (88) have shown that connective tissue and mesodermal derivatives are inhibited selectively and the adrenocortical hormones have been shown to interfere with the growth of the chick embryo, mouse and rat, and of lymphatic tissue, skin and hair, and of certain transplanted mouse and rat tumors (52, 112). Why certain normal and neoplastic tissues are susceptible to the action of these hormones is not known.

Specific Cell Poisons

RADIOACTIVE ISOTOPES

Radioactive isotopes are considered as falling within the group of chemotherapeutic agents because their chemical properties, to a large extent, permit them to localize in specific tissues, where they can exert their destructive effects through radiation. Of the large number of radioactive isotopes available for study, two have found a definite place in the treatment of cancer: radioactive phosphorus (P^{32}) and radioactive iodine (I^{131})

Radioactive Phosphorus

P^{32} was one of the first radioactive isotopes used clinically It produces beta rays; its physical half-life is 14 3 days, and its biological half-life in man is about 8 days

P^{32} may be given orally or intravenously The oral route is preferred in our clinic (28). P^{32} is taken in water just before breakfast, an empty stomach may facilitate its absorption The usual dose is 1 millicurie per 10 kg of body weight, divided into 5 daily doses, so that ⅕ the total dose, about 1.4 millicurie in the average adult, is taken each day While this is an average dose, it may be excessive in some cases and insufficient in others. The therapeutic and bone marrow depressant effects of P^{32} may take 4 to 6 weeks and sometimes longer to develop, and the decision concerning further therapy demands a proper respect for this interval. The limiting factor of

and this followed a severe infection, and two children obtained some degree of symptomatic improvement. While the survival of these patients was somewhat prolonged over that of the untreated cases, 50 per cent of the patients died within 7 months and 95 per cent were dead at the end of the first year.

Brief, spontaneous remissions in acute leukemia have been noted by many observers Diamond is said (32) to have found spontaneous remissions in 10 per cent of 300 children with acute leukemia seen at the Children's Hospital in Boston, and these remissions were frequently associated with severe infections. Four per cent complete spontaneous remissions and 8 7 per cent symptomatic remissions have been reported by Southam et al. (107). The spontaneous remission usually occurs only once during the course of the disease, but Rosenthal has discussed (32) a patient who had six such remissions on treatment with blood transfusions and survived for 13 months. It may be concluded that acute leukemia is a rapidly progressive disease, which, in children, may undergo temporary spontaneous remissions in 5 to 10 per cent of the cases; supportive therapy may cause some prolongation of life but does not affect the course of the disease, and a small percentage of children may have subacute or chronic types of leukemia, and may survive one to two years or longer. There has been no evidence that x-ray therapy, radioactive phosphorus, urethane or nitrogen mustard caused a consistent number of hematological remissions in these patients, or appreciably prolonged life.

In 1948, Farber and his co-workers (33) reported on the effects of 4-amino PGA (Aminopterin) on the course of acute leukemia in children Of 16 patients treated, 10 showed some type of favorable response and 6 failed to improve In the responsive cases, the white cell count tended to return to normal levels and the percentage of immature cells found in the peripheral blood and bone marrow reverted toward normal Furthermore, there was an increase in the hemoglobin level, red cell and platelet counts, as further evidences of hematological remission This original report emphasized the temporary nature of these remissions, and correctly anticipated the results that were obtained subsequently with the 4-amino derivatives of folic acid in a large series of patients with acute leukemia Farber's observation was promptly extended in a number of other institutions and there have been numerous reports on the subject. The methods of analyzing the results of treatment in different clinics are not entirely uniform, but the data obtained in large independent series have been fairly consistent In the most recent report by Farber and Downing (34), a total of 194 patients was treated initially with a folic acid antagonist, 62 8 per cent of the cases showed an important degree of improvement, in 9 3 per cent of the series the remissions were associated with infections, 25 per cent of the total group showed

of their disease. The physician has the problem of managing these patients and applying appropriate forms of treatment. When presented with such a patient, the physician should review carefully the clinical history and previous treatment, and obtain unquestionable evidence, and a repeat biopsy, if necessary, to prove that the patient has cancer and is beyond the possibility of any curative procedure. In inoperable cancer it must be remembered that palliative surgery or x-ray therapy may be of great value, and that encouragement and general supportive measures alone have sustained patients for significant periods. In certain situations the chemotherapeutic agents already described have contributed substantially to the comfort of the patient, and, in some instances, they have prolonged life. The indications for, and the effects of, these drugs in various types of neoplastic disease will be discussed in the remainder of this chapter.

Acute Leukemia

Acute leukemia in children is a fatal disease which usually runs its course in 3 to 8 months. Southam et al. (107) reviewed the natural history of acute leukemia in 172 patients, most of the patients died within 6 months and only 2 patients survived longer than one year. There was little difference in survival time in patients with cell types classified as myeloid or lymphatic in origin, or in whom the white cell count was high or low at the onset of the disease. In a review of 158 children with acute leukemia, Rodgers et al. (92) reported a male:female ratio of 2:1 and the average age at onset of 4 years and 11 months. The cases were classified as lymphatic 133, granulocytic 12, monocytic 6, and 7 were stem cell leukemias. In 152 patients followed, the average duration of life from onset of symptoms to death was 4.2 months. One hundred and thirty-nine cases were dead within 9 months with an average survival of 2.9 months. Eleven cases had an average survival of 12.4 months and these were called subacute leukemia; two chronic granulocytic leukemias survived for 50 and 53 months, respectively. Bierman et al. (5) analyzed the survival time in 76 children with acute lymphatic leukemia. The disease began at an average age of 5.0 years and the average survival of 59 children, either untreated or treated with x-ray therapy or blood transfusions, was 5.6 to 6.0 months. The 17 remaining patients, *who received blood transfusions and antibiotics*, survived an average of 8.9 months and 9 of these patients survived for 9 to 23 months. The writers suggested that supportive therapy may produce a significant prolongation *of life, which must be taken into* account in assessing the therapeutic value of new agents. Burchenal (8) reported on 19 children with acute leukemia treated with a nitrogen mustard derivative (SK 136) (table 2), together with supportive measures as indicated. A complete, temporary hematological remission occurred in one child only,

porarily to permit recovery. After several episodes of toxicity an occasional patient will suddenly go into remission, but in about 50 per cent of the cases no improvement ever occurs. In these patients, and in the previously responsive patients who have become refractory to treatment, the active leukemic process may clinically resemble A-methopterin toxicity, as seen from the leukopenia, anemia, thrombocytopenia, petechiae, gross bleeding and fever, and it is often difficult to decide whether to stop therapy in the hope that the child will improve as toxic manifestations clear, or to continue it in an attempt to control the leukemia process. These situations require considerable judgment and intuition.

Figure 3 illustrates the course of the disease in a 2-year-old girl who temporarily responded to anti-folic therapy.

Within one year after the effects of the folic acid antagonists were reported, cortisone and ACTH became available and these agents were found to induce temporary remissions in acute leukemia (83). Symptomatic improvement occurs within a few days, with an increase in general well-being, and bleeding manifestations and fever disappear. Hematological improvement often occurs rapidly with a return of the white cell count to normal levels, a rise in platelet count and a reticulocytosis, the bone marrow shows a marked increase in erythropoiesis, followed by a decrease in the blast forms and appearance of the normal myeloid series. The remissions usually are of short duration, from 3 weeks to 2 months, and during relapse further treatment sometimes induces second and third remissions, the patient, however, finally becomes refractory to further treatment. ACTH and cortisone have induced remissions in 50 to 60 per cent of children treated, Schulman et al. (98) obtained 6 complete and 2 partial responses in 14 patients, and Darte et al (25) reported temporary remissions in 50 per cent of his cases. Pierce, in table 5, has summarized her results with ACTH in the treatment of acute leukemia in children.

If the child is desperately ill, a continuous drip of intravenous ACTH may be most rapidly effective but, for routine use, oral cortisone is more convenient. If the patient shows a response, treatment should be continued until there is bone marrow evidence of complete hematological remission. During vigorous treatment the child may develop the undesirable effects of excessive adrenal cortical activity, with increased appetite and weight gain, fluid retention and edema, hypertension, hypertensive encephalopathy with episodes of coma, and mental changes. The dose may have to be decreased or treatment stopped. When a complete remission has occurred, it is usually desirable to discontinue therapy but if the child is not responding satisfactorily treatment should be continued for at least 3 weeks, and the dosage raised until definite signs of excessive adrenal cortical activity result. These patients occasionally may show increased susceptibility to

complete clinical and hematological remissions, and 37.2 per cent of the patients failed to respond. Burchenal et al. (10) carefully analyzed the clinical course of 60 children with acute leukemia treated with 4-amino PGA and derivatives; 31.7 per cent of the group obtained complete clinical and hematological remissions. Partial remissions were described in only 5 per cent of the cases. *Treatment usually was stopped when the patient* went into remission, and these periods of hematological improvement usually lasted for 5 to 8 weeks. Patients responding to the first course could be expected to respond several more times, but they eventually become resistant to treatment. The mean survival time, from the onset of therapy, in the responsive cases was 14.7 months (6 to 24 month range), as compared to 2.1 months (1 day to 7½ months) in the failures. Fourteen children of the 60 treated (23 per cent) survived longer than one year after the onset of treatment. The Second Conference on the Folic Acid Antagonists held in Boston in March, 1951 (32) summarized the results obtained by various clinics; the data were in good agreement and consistent with the above observations.

The following procedure is used in our clinic for the treatment of a proved case of acute leukemia with folic acid antagonist therapy. The drug of choice is A-methopterin, and it is begun at a daily dose of 2.5 mg in the average child, smaller doses are given to infants. The patient is followed closely for evidences of mouth lesions, rapid fall in white cell count or symptoms of diarrhea or abdominal cramps. If any signs of toxicity appear the drug is stopped temporarily but if, at the end of 10 days, no evidence of toxicity or clinical improvement appears, the dose is increased to 5 mg The increase in dosage may precipitate toxic symptoms, and the child must be watched carefully. In Farber's series 65 per cent of the remissions occurred without toxicity, in Burchenal's series, only 42 *per cent showed no evidence of toxicity* Harbingers of remission may be a fall in the white cell count, improvement in the white cell differential, rise in platelet and reticulocyte counts, fall in fever and the maintenance of hemoglobin level without blood transfusions, but the only reliable criterion *is a decrease in the blast forms and an increase in maturing granu*locytes and erythroid elements in the bone marrow During active treatment, weekly bone marrow aspirations are taken as a guide to further treatment If marked improvement is seen, A-methopterin therapy is discontinued, *and the patient watched carefully for beginning signs* of relapse, which will be detected first in the bone marrow In some clinics A-methopterin is continued as maintenance therapy in order to prolong the remission There is no proof as to whether continuous or intermittent therapy is

infection, and if a severe infection is present, treatment should be discontinued if possible. In desperate situations, treatment has been continued in the presence of hypertension or infection, without necessarily aggravating the complication, and sometimes a hematological remission has resulted In general, ACTH and cortisone alone have not been shown to prolong life in leukemia significantly

Excessive adrenal cortical activity and the folic acid antagonists act by different mechanisms on the leukemic process, and patients refractory to cortisone may respond to the anti-folics and vice versa Both agents are used, therefore, for their independent effects in acute leukemia If the child, when first seen, is in poor general condition, with hemorrhagic tendencies,

TABLE 5

Remission Rate Obtained with Successive Courses of ACTH in 37 Cases of Acute Leukemia in Children (87)

COURSE OF ACTH	NUMBER OF CASES TREATED	NUMBER OF CASES WITH REMISSIONS	REMISSION RATE	DURATION OF REMISSION	
				Average	Range
			%	wks	wks
1st	37*	28	75 67	6 5	2–16
2nd	23	12	52 50	6 2	2–12
3rd	15	6	40 00	4 6	2–7
4th	6	2	33 33	5 5	5–6
5th	2	1	50 00	5 0	
6th	1	0	0		

* 8 cases treated previously with Aminopterin, 6 cases living after only one course of ACTH

an elevated white cell count, and there is the possibility that he may not survive long enough for an adequate trial of A-methopterin, ACTH or cortisone is indicated. In some clinics, A-methopterin is started at the same time, so that if the patient does respond to cortisone, the remission may be prolonged by the anti-folic, and if he is unresponsive to excessive adrenal cortical activity, some progress has already been made toward inducing a "folic acid" deficiency. Our practice is to start ACTH or cortisone in the very sick patient and continue it until a definite remission, or maximal improvement is obtained. Then, A-methopterin is started, and continued during the usual duration of the cortisone remission and if the patient remains in remission it is discontinued. The patient is watched carefully and at the first sign of bone marrow relapse A-methopterin is re-started and used in the conventional fashion It is employed as long as the child is responsive, but when he becomes refractory to the anti-folics despite the the production of toxicity, cortisone or ACTH is used again.

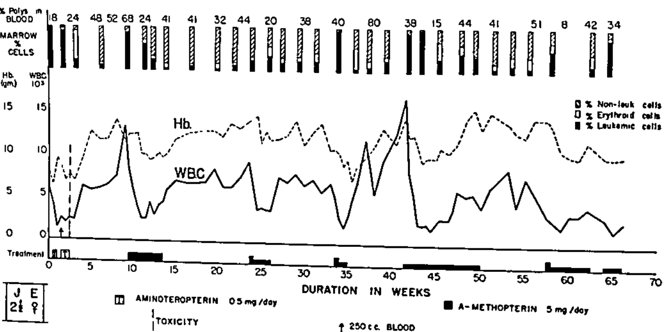


FIG 3 J E, a 2 year-old female, demonstrated an initial remission within three weeks after the onset of treatment, as shown by a decrease in leukemic cells in the marrow and return of erythropoiesis The patient was initially treated with Aminopterin but relapsed after 10 weeks, and was subsequently treated with A-methopterin. She obtained 3 complete remissions during the first 60 weeks of treatment, as shown on the chart; subsequently two more remissions occurred with A-methopterin, but in the 77th week of treatment, when it was apparent that the patient had become refractory to the folic acid antagonists, ACTH was started ACTH and then cortisone caused brief symptomatic improvement, without change in the bone marrow, and she died 86 weeks after the onset of therapy, and 90 weeks after the clinical onset of the disease

factory method of controlling it. If the disease is bulky and encroaching on important structures, a course of HN2 or TEM may produce rapid involution of the disease, and then x-ray therapy can be given to the resistant areas. A trial of the folic acid antagonists is advisable in the widespread disease and, finally, ACTH or cortisone may be of temporary value

Adults with acute leukemia are less responsive to treatment than are children. In 28 adults reported (10) only one patient (3 6 per cent) showed a complete hematological remission on anti-folic therapy. This was a remarkable case; a 23-year-old girl with a confirmed diagnosis of acute leukemia, who went into clinical and hematological remission on 2,6-diaminopurine and amino-an-fol and remained entirely well and without evidence of disease for 3 years when, for the first time, evidence of bone marrow relapse appeared. Stickney (32) treated 40 adults with acute leukemia with the anti-folics, and obtained complete remissions in 3 cases (7.5 per cent); Bethell (32) reported the best results to date, with 16 "good" responses, 13 "fair" responses and 27 "poor" responses in 56 patients with various types of acute leukemia A "good" response, described as a patient showing "substantial clinical and hematological improvement, temporary cessation of hemorrhagic manifestations when these were present, decrease in the number of transfusions required and apparent prolongation of life" occurred in patients less than 35 years of age A "good" response is not necessarily a complete hematological remission. It seems reasonable to conclude that young adults may show a partial response to the anti-folics, and possibly some prolongation of life and, for this reason, a trial of the anti-folics may be justified in patients under 30 years of age, in the older patients the results have been too poor to merit the widespread use of the anti-folics

ACTH and cortisone have induced brief and striking hematological remissions in about 20 to 25 per cent of young adults treated, and symptomatic improvement may occur in perhaps another 20 per cent of the patients The remissions, when they occur, may develop as rapidly and are as immediately satisfactory as those seen in children, but relapses occur precipitously and second remissions are induced rarely A trial of either intravenous ACTH or oral cortisone, depending on the clinical condition of the patient, is suggested in the young adult with acute leukemia, along with general supportive measures

Nitrogen mustard, TEM or x-ray therapy generally has been found to be without value in acute leukemia Occasionally, however, a patient may be classified hematologically as acute leukemia, but the spleen is enlarged, and bleeding manifestations and thrombocytopenia are not present. Symptomatic improvement may sometimes result from x-ray therapy to the spleen or a cautious trial of TEM Monocytic leukemia has not responded

In children in relatively good condition at the outset, the anti-folics are given initially since if a remission occurs it is more satisfactory and prolonged than that following ACTH or cortisone. If the child proves to be unresponsive to an adequate course of A-methopterin, cortisone is given Terminally, in the anti-folic and cortisone-resistant patient, a trial of intravenous ACTH has sometimes induced a remission. By the manipulation of the hormonal and anti-metabolite therapy more satisfactory results are obtained than from either agent alone. *Kingsley-Pillers et al.* (65) reported in detail on the alternation of ACTH, cortisone and the folic acid antagonists in treating leukemia. Sixteen children, resistant to the anti-folics, were treated with adrenocortical hormones, and 10 showed good clinical and hematological responses, one a partial response, 2 showed some evidence of bone marrow response but died during treatment, and 3 failed to improve. Six patients, refractory to adrenocortical hormone therapy, were treated with A-methopterin and 3 showed a good clinical and hematological response, one a partial response, and two failed to improve. It is estimated that about 50 per cent of the patients that become resistant to one form of therapy will respond to the other types Pierce (87) has found that when ACTH or the anti-folics were given alone only 8 6 per cent and 8 0 per cent, respectively, of the children with acute leukemia were alive at the end of one year, whereas with both agents available, 28.6 per cent were living at that time

It should be emphasized again that during the course of acute leukemia supportive measures should be used in conjunction with specific chemotherapy. Blood transfusions and antibiotics are given as indicated, and, if the patient has extensive local involvement with the disease, small doses of localized x-rays may give satisfactory relief.

While the immediate response to chemotherapy in acute leukemia may be spectacular, in viewing the end-results, the great effort involved has not seemed very rewarding, and some clinics have questioned the value of treating this condition It is our strong belief, however, that these children should be given the opportunity of a proper trial of chemotherapy, both because of the important improvement that may result, which is gratifying to the patient and family, and because of the possibility that in prolonging their lives, some of these patients may survive *long enough to benefit* from the discoveries of more effective methods of treatment.

Lymphosarcoma in children may appear first as a local disease involving the node-bearing areas, liver and spleen but, during the later stages, the disease frequently disseminates to give a picture very similar to that of acute leukemia. The anti-folics may induce regressions in lymphosarcoma in children as striking and as frequent as those occurring in acute leukemia. In patients with localized disease x-ray therapy may be the most *satis-*

an attempt at cure, and the limited objectives of treatment are to temporarily control the disease process and to relieve symptoms. There are several procedures which may keep the patient for a time in good general health and enable him to engage in normal activities during the major period of his disease There has been, however, no substantial evidence that present forms of treatment cause a statistically significant prolongation of life as compared to untreated cases, although in special situations treatment may be life-saving and, thus, temporarily prolong survival time The available therapy for the chronic lymphomas and leukemias appears to be largely suppressive; the manifestations of the disease but not the underlying process are controlled temporarily, and the latter continues to progress in its predestined fashion This view of the natural course of the lymphomas and the effects of therapy, is shown graphically in figure 4. This illustrates a rapidly progressive, intermediate and slowly progressive form of lymphoma, and the effect of therapy in temporarily restoring the patient to almost normal health for varying periods of time, depending on the rate of progression of the disease. The baseline, however, shows the fundamental pattern of the disease

Chronic Myelogenous Leukemia

Chronic myelogenous leukemia usually runs a prolonged course during which the patient responds satisfactorily to several different types of therapy, and then a rapidly progressive terminal phase appears, during which resistance to all types of treatment develops Data summarized from several clinics by Diamond and Craver (27) show an average survival time from the clinical onset of the disease of 3 to 3.6 years Shimkin et al (103) reviewed the course of 212 patients with chronic myelogenous leukemia seen in San Francisco during 1910 to 1948, and compared these data with a total of 975 patients reported in the literature They conclude that the survival time has shown no significant change in the past 30 years, the mean survival time is 3 to 4 years, with 20 per cent of the patients living 5 years or longer and 2 per cent living 10 years or longer The therapeutic measures that are available will suppress the manifestations of the disease and sustain the patient in relatively good health most of the time, but apparently will not affect its progression The choice of a method of therapy is based largely on the experience of the physician, its ease of administration and convenience to the patient

Radiation therapy, P^{32}, HN2, TEM and allied compounds, urethane, potassium arsenite and benzene and, less satisfactorily or consistently, the folic acid antagonists and 2,6-diaminopurine have induced clinical remissions The manifestations of improvement consist in a reduction in the size of the spleen, a fall in the white cell count, improvement in the differen-

with any regularity to any chemotherapeutic agent, including nitrogen mustard, urethane, P^{32}, ACTH or cortisone or the folic acid antagonists, but there have been isolated reports of improvement in the bone marrow picture. The course of acute leukemia is discouraging to the patient and physician, and it is entirely justifiable to try some form of therapy in order to sustain the patient's morale, and because of the rare possibility of inducing a hematological remission.

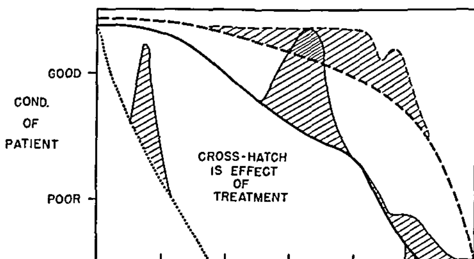


FIG 4 SCHEMATIC REPRESENTATION OF THE NATURAL COURSE OF DISSEMINATED LYMPHOMAS AND CHRONIC LEUKEMIAS

Chronic Leukemia and Lymphomas

The chronic leukemias and lymphomas appear to run their individual courses in a manner apparently predetermined, to a large extent, in each patient at the outset of the disease There is no definite evidence of specific host factors which have influenced significantly the natural evolution of the neoplastic process In some cases the disease may proceed at a rapid rate, death occurring within a few months after the clinical onset, whereas in other cases it may run a fairly benign course for many years, and show acceleration only in the final stages In rare instances lymphosarcoma and Hodgkin's disease may remain localized, and local excision and/or intensive radiotherapy have been curative. These cures have occurred particularly in lymphomas in the *cervical region, reticulum cell sarcomas of* bone and lymphosarcoma of the gastrointestinal tract In the vast majority of cases, however, the disease is disseminated too widely when first seen to permit

and an elevated blood urea or uric acid, treatment should be given slowly in order to decrease these renal complications

URETHANE

This agent is effective in producing remissions; the usual dose of 2 to 4 grams daily given within one to three weeks will cause, in most cases, a gradual decrease in the white cell count, rise in hemoglobin level and reduction in the size of the spleen By the continuous and carefully controlled administration of urethane, the patient may be maintained in remission for long periods, but he ultimately becomes refractory to the drug (50, 82).

FOWLER'S SOLUTION

Potassium arsenite is a simple and effective treatment for the early stages of the disease Proper dosage will bring the white cell count down and result in a rise in hemoglobin without the hazard of serious depression of the bone marrow In the more advanced stages of the disease other agents are more effective

The remissions induced by these agents in time become briefer and less complete, and the patient finally goes into the terminal stage; usually with a rising white cell count, anemia, thrombocytopenia, enlarging spleen, liver and lymph nodes, bone pain and tenderness, and a more acute bone marrow picture Intensive use of a chemotherapeutic agent may produce a transient fall in the white count but it promptly rises. The shift from one agent to another may occasionally be of value, and, if the liver and spleen have enlarged rapidly, x-ray therapy to these organs may be of considerable benefit Although the evidence of accelerated activity of the disease is ominous, persistent and intensive therapy may salvage a patient temporarily.

Chronic Lymphatic Leukemia

Chronic lymphatic leukemia has a variable, but often a fairly benign course during the early stages of the disease An elevated white cell count, an enlarged liver, spleen and lymph nodes, may be found in patients in relatively good general condition, and these patients are sometimes best treated by observation only If the chief difficulty is a slowly developing anemia and weakness, periodic blood transfusions may relieve these symp- [illegible] chemotherapeutic agents, and excessive dosage may result in prolonged and serious depression of hematopoietic activity. The average duration of life in chronic lymphatic leukemia from the onset of symptoms, is in the range of 3 to 4 years (28).

tial count, a spontaneous rise in the hemoglobin level and an increased feeling of well-being. In the early, relatively asymptomatic stage of the disease treatment should be deferred until a specific indication occurs, such as anemia, increasing fatiguability and a rise in the white cell count. The most useful methods of producing improvement, not necessarily in order of effectiveness are:

X-RAY THERAPY TO THE SPLEEN

A total dose of 400 to 600 r. delivered over the spleen in 4 to 10 treatments may cause a marked shrinkage in the spleen and clinical and hematological improvement. Remissions may last for several months or longer, and treatment is restarted if the white cell count rises or symptoms recur.

RADIOACTIVE PHOSPHORUS

A course of P^{32} will induce a gradual fall in the white count, and a rise in hemoglobin, but splenic regression may not be so striking as following local x-ray therapy to the spleen The usual dose of 0.1 millicurie per kg may not be adequate and in 6 weeks, if a satisfactory response has not occurred, further treatment may be given The usual remission lasts about 4 to 6 months, and the patient may be "retreated" when signs of relapse appear. In cases where splenomegaly is a serious problem, x-ray therapy to the spleen gives a more satisfactory result.

NITROGEN MUSTARD AND TEM

HN2 will induce a satisfactory remission which usually continues for one to 4 months, or an average of two months (12). In some patients this treatment may be given repeatedly; but the short remissions and the nausea and vomiting induced by HN2 make it less desirable than other forms of therapy In patients with far-advanced leukemia no longer responsive to other forms of therapy, a trial of HN2 or TEM which brings the white cell count down to low levels may sometimes induce another hematological remission. Oral TEM has proved to be effective and useful in patients with chronic myelogenous leukemia TEM may be given at weekly intervals until the white cell count has returned to normal levels, and maintenance doses can then be continued An occasional complication of intensive and rapid therapy has been oliguria (and rarely polyuria), sometimes occurring with hematuria, and uremia and hyperuricemia This is apparently due to the rapid breakdown of tissue with formation of large amounts of uric acid, which may be directly injurious to the kidneys, particularly if they are already impaired by the underlying disease Kravitz et al. (67) have reported a patient, treated with TEM, who developed acute hyperuricemia and uremia, and then recovered In patients with a high white cell count

URETHANE

Although some patients have responded to urethane (50, 82) this agent does not appear to be of any special value in chronic lymphatic leukemia.

ACTH AND CORTISONE

ACTH and cortisone, which have little effect on chronic myelogenous leukemia, may be of considerable value in chronic lymphatic leukemia Of 8 patients treated by Pearson and Eliel (83) all showed some shrinkage of enlarged lymph nodes, liver and spleen, and in 5 patients there was a spontaneous rise in hemoglobin levels, and these levels were maintained for longer periods following blood transfusions. It was of interest that all patients initially showed a rise in the lymphocyte count in the peripheral blood, and this was followed by a fall in count. None of the patients showed any improvement in the differential count of the peripheral blood, or in the bone marrow. In the hemolytic anemias associated with chronic lymphatic leukemia, adrenal steroid therapy may be of great value (93).

In general, the conservative management of lymphatic leukemia with supportive measures and localized x-ray therapy has proved most satisfactory. In our experience, oral TEM may be useful as a systemic agent, and it occasionally will produce clinical and hematological remissions Radioactive phosphorus, nitrogen mustard and urethane are not as convenient to use In the far-advanced stages of the disease, a trial of cortisone or ACTH may result in temporary improvement

Hodgkin's Disease

It is widely held that Hodgkin's disease is a variant of the lymphoma group Although the involved tissues may present a characteristic histological appearance, in some instances the same patient may have simultaneous biopsies diagnosed as Hodgkin's disease and lymphosarcoma or, during the course of the disease, repeated biopsies may be interpreted differently The classification of Custer clearly indicates the inter-relationships of the lymphomas, and this would appear to be the most satisfactory working interpretation of these conditions, until such time as specific etiological factors or responses to a specific therapeutic agent can be used to distinguish the different forms of lymphoma

As already noted, Hodgkin's disease runs a variable course, and the proper treatment of the disease will depend on the clinical stage and manifestations of the disease This is shown graphically in figure 4. In Craver's (19) series of patients, in 265 cases seen between 1918–1935, 17.7 per cent survived for 5 years, whereas in 471 cases seen during 1930–43, 23 6 per cent were living at the end of 5 years These results suggest, but do not prove, that treatment may have produced a modest increase in survival

Under proper management, these patients may remain in relatively good condition during the major portion of their disease, and they usually succumb to either inter-current disease, secondary infections, or progressive and severe anemia with thrombocytopenia and hemorrhagic manifestations

The several modes of treatment available are applied more cautiously and less vigorously than in chronic myelogenous leukemia.

X-RAY THERAPY

This may be given to localized masses which are producing symptoms such as enlarged peripheral nodes or abdominal nodes. A moderate dose of 200 to 400 r. is used since some of the nodes may be radiosensitive; if these do not respond it may be necessary to use larger doses.

RADIOACTIVE PHOSPHORUS

P^{32} has been recommended as a convenient form of systemic therapy. This treatment will reduce the peripheral blood count, but improvement in the hemoglobin level, or substantial regression of enlarged nodes is an infrequent occurrence, and it may be necessary to apply x-ray therapy to resistant areas

NITROGEN MUSTARD AND TEM

The usual course of 0.4 mg. per kg. of HN2 may be excessive, and the patient should receive 0 1 to 0 2 mg per kg, and further treatment considered after 1 to 2 weeks if the white cell count has not fallen. The objectives of therapy, more limited than in chronic myelogenous leukemia, are to produce some fall in the white cell count, a partial reduction of enlarged nodes and organs, and a rise in hemoglobin levels The duration of the improvement depends on the rapidity with which the disease is progressing, and some patients have been appreciably improved for several years (12, 109)

Oral TEM has been more generally useful than HN2. By giving the agent cautiously in doses of 2 5 to 5 mg initially, the white count has been reverted to normal in some, and there has been marked and prolonged clinical improvement (57, 96, 104) Such a response is described in the following patient

A S, a 62 year-old man was found in March, 1949 to have a generalized lymphadenopathy, enlarged liver and spleen, and white cell count of 300,000, composed almost entirely on lymphocytes During July to November, 1949 he received a total of 12 mg TEM intravenously, with a fall in the white cell count and a rise in hemoglobin level In March, 1950, 6 months later, the white cell count began to rise, and 5 mg of TEM orally reduced it to normal levels This dose was repeated 2½ months later and, at present, 2 years after the last dose, the patient remains in excellent health, with a normal bone marrow and with no physical evidence of disease

It is unreasonable, however, to use large doses of x-ray therapy in an attempt to obliterate generalized disease. HN2 and TEM have been used successfully to control generalized early disease, but they are not as generally useful or as locally effective as x-rays.

GENERALIZED, SYMPTOMATIC DISEASE; PATIENT IN GOOD GENERAL CONDITION

When the disease is generalized the patient may complain of itching, anorexia, fever, weakness and weight loss, and present lymphadenopathy, local areas of disease infiltration and an enlarged spleen and liver. These patients should be treated adequately and persistently. In the ambulatory patient oral TEM may be very useful in relieving and temporarily controlling many of the symptoms, producing regression of enlarged organs and restoring the patient to normal activity. TEM is administered as described

TABLE 6

EVIDENCE OF DISEASE	NO OF PATIENTS	NO IMPROVED
Fever	15	13
Itching .	14	14
Weakness	16	13
Enlarged nodes	16	13
Palpable liver	4	3
Palpable spleen	2	2
Pulmonary symptoms, cough, dyspnea, etc	12	11
Abnormal chest roentgenogram	12	5

in table 1 The usual course of treatment may take one month, and further TEM is withheld when the white cell count has shown a definite fall or a satisfactory clinical response has been obtained. As soon as there is evidence of relapse, treatment is restarted, and in some patients maintenance therapy is successful In our series of 25 patients treated with oral TEM, 20 were improved, and the evidences of disease were relieved temporarily, as shown in table 6

The average duration of improvement was 8 weeks (2 to 14 weeks) and 8 of 13 patients responded to a second course of TEM While there may be an excellent systemic response to TEM, areas of local disease may persist and actually increase in size. These areas should be treated with x-ray therapy Bone lesions have been particularly resistant to HN2 or TEM therapy, and x-ray therapy should be instituted as soon as definite bone pain occurs or an osseous lesion is diagnosed by roentgen examination.

An alternate form of chemotherapy is intravenous nitrogen mustard (HN2) This agent is usually given to hospitalized patients but, in some cases, it can be used in the Out-Patient Department. It has about the

time. Gellhorn and Collins (41), reviewing the results obtained from the use of nitrogen mustard in addition to x-ray therapy in the management of Hodgkin's disease, have concluded that life has not been prolonged appreciably, although palliation has been improved. Irrespective of whether convincing statistical evidence of prolongation of life for the entire group of patients can be obtained, each patient should be followed carefully and receive appropriate and adequate therapy during the course of the disease.

LOCALIZED DISEASE

Hodgkin's disease may first present itself with enlargement of the cervical, axillary and mediastinal nodes with minor or absent systemic symptoms. While the disease is usually disseminated when first seen and beyond the hope of cure, the fact that evidence of disease can be found only in these areas makes it tempting to try to obliterate the process. When the disease appears confined to the cervical or axillary areas local resection together, in some cases, with extensive radiotherapy has resulted in prolonged remissions and possibly cures. It is, of course, impossible to be certain that the favorable course in a particular patient was due to treatment since, untreated, patients have sometimes run a long course without apparent progression of the disease. When Hodgkin's disease is too extensive to be resected, but still apparently confined to a circumscribed area, it has been treated by intensive x-ray therapy. While it is apparent that these patients may have occult disease elsewhere and the most vigorous attempts at local therapy will thus be doomed to failure, it would, nevertheless, be unfortunate if, by withholding radical therapy, these patients were deprived of an opportunity for prolonged remission or cure. It has been our practice, therefore, to treat circumscribed but inoperable disease with intensive x-ray therapy, and recently a combination of nitrogen mustard and x-ray therapy has been used. Our procedure consists in giving 0.4 mg per kg. of HN2 intravenously (with tourniquets applied to both legs and one arm for 5 minutes), and a series of x-ray treatments is immediately started to the involved area. In order to try to obtain the additive effects of HN2 and radiation, the course of x-ray therapy, which has run up to 2000 to 3000 r. tumor dose, has been delivered within 8 days following the injection of HN2. Another justification for the use of HN2 is the possibility that it may have an important effect on undetectable small foci of disease elsewhere in the body This treatment induces a transient and moderate to marked leukopenia 7 to 10 days after the injection of HN2 and symptoms due to the x-rays, such as esophagitis and tracheitis may be present for several weeks or longer.

When enlarged nodes are present in several areas, and the patient is in good general condition, these nodes should be treated by localized radiation

some extent by nitrogen mustard therapy. When severe bone marrow depression occurs nitrogen mustard therapy is no longer feasible and, in most cases, x-ray therapy is also ineffective. A trial of cortisone or ACTH may temporarily cause symptomatic improvement, and it is worth a trial.

FAR-ADVANCED DISEASE

A course of HN2 or TEM is indicated in every patient with far-advanced, widespread Hodgkin's disease, particularly in whom x-ray therapy is no longer considered feasible. In these debilitated patients, HN2 or TEM may be given intravenously and it may be justifiable to treat the patient to the point where a severe depression of the marrow is induced in a desperate attempt to produce a remission in an otherwise hopeless situation. While it is recognized that nitrogen mustard may further depress an already impaired bone marrow, occasionally clinical improvement and a rise in hematopoietic activity above pre-treatment levels has occurred.

ACTH or cortisone is occasionally useful in inducing symptomatic improvement but they have not altered appreciably the progress of the disease. In a group of 10 patients reported by Strauss et al. (114) 7 obtained some transient subjective improvement, 3 patients showed a fall in fever, and one showed definite tumor regression, and some degree of stimulation was noted in the maturation of the bone marrow. In our experience, the severe bone marrow depression occurring in Hodgkin's disease has not been appreciably improved by ACTH or cortisone. In the occasional cases of hemolytic anemia, however, cortisone may be of real value. Other agents, such as urethane, radioactive phosphorus and the folic acid antagonists have not been effective in Hodgkin's disease, in our experience.

In general, radiation therapy and nitrogen mustard and allied compounds maintain patients with Hodgkin's disease in an improved condition during the course of the disease and, in some cases, treatment may possibly cause a modest prolongation of life Nitrogen mustard has induced periods of improvement in patients thought to be unresponsive to further x-ray therapy (23). Supportive treatment is an essential part of the management of the disease. In view of the considerable degree of unpredictability of the course of Hodgkin's disease and its response to treatment, agressive and persistent therapy is indicated in all patients.

Lymphosarcoma

The pathological diagnosis of lymphosarcoma comprises a variety of interrelated conditions, including giant follicular lymphosarcoma, small cell lymphosarcoma (which is distinguished from lymphatic leukemia by the absence of bone marrow and peripheral blood involvement), leukolymphosarcoma, which shows some bone marrow involvement but with little change

same degree of effectiveness as oral TEM but the latter has advantages in that it does not induce nausea and vomiting as frequently, and the maximum tolerated dose can be more easily approximated by protracted administration. In hospitalized patients in whom rapid treatment is desirable, intravenous HN2 is indicated. A preparation of intravenous TEM is now available, which rarely induces any gastrointestinal symptoms at single doses of 0.04 mg per kg., a dose which has approximately the same activity as 0.1 mg. per kg of HN2. Intravenous TEM may in time, therefore, replace intravenous HN2 in many situations. However, in cases in which one wishes to administer a full course of treatment in a single dose, HN2 is used in preference to TEM. At these large doses TEM also causes nausea and vomiting, which may be more persistent than with HN2 and, because of greater experience, the maximum tolerated single dose of HN2 can be estimated more safely.

Patients with serious symptoms due to local extension of Hodgkin's disease—such as compression of the cord with paraplegia, pressure on the trachea, or partial occlusion of great vessels—may be rapidly relieved by a course of HN2 (0.4 mg per kg) given in a single injection. This has not induced edema in the tumor and, following the temporary regression induced by HN2, x-ray therapy can be delivered to the local disease

LOCAL EXTENSIVE DISEASE

One type of Hodgkin's disease may persist to a large extent in a single area, extending locally to produce serious complications, pulmonary infiltration, skin nodules with ulceration and bone involvement are examples While these lesions may be extremely resistant to x-ray therapy, its vigorous use offers the best possibility of controlling these processes. Sometimes a course of HN2 or TEM may induce partial and temporary regression of the disease and relieve some of the distressing symptoms If improvement follows HN2 this would indicate that the disease is still responsive to therapy, and further x-ray therapy, if feasible, should be tried

SYSTEMIC SYMPTOMS WITH LITTLE DEMONSTRABLE EVIDENCE OF DISEASE

In another type of Hodgkin's disease, severe systemic symptoms such as high fever, sometimes of the Pel-Ebstein type, itching, weakness and anemia may predominate, with minimal or absent palpable disease These patients may respond remarkably to a course of HN2 or TEM, with a fall in fever, increase in appetite and weight, and a restored feeling of well-being These remissions may last for a few days to several months, depending on the rate of progression of the disease, but the patient finally becomes refractory to treatment The fever continues to rise and persist, while leukopenia, thrombocytopenia and anemia become more severe, possibly aggravated to

that it can not be controlled by x-rays. In rare instances, ACTH or cortisone, given in large doses, has had a temporary favorable influence on the disease. Five patients with lymphosarcoma showed some regression of disease on ACTH but, despite maintenance therapy, all relapsed within 3 to 4 months (93). Two of the patients had hemolytic anemia and they were strikingly benefitted

Bone marrow depression with anemia, thrombocytopenia and bleeding may be a conspicuous finding in far-advanced lymphosarcoma, and supportive measures with blood transfusions and antibiotics are of value. The folic acid antagonists, P^{32} and urethane have not proved of practical value in these conditions in adults.

Mycosis Fungoides

Mycosis fungoides, while showing predominantly skin lesions, appears to be a variant of the lymphomas. In the early stages of the disease, and when the skin lesions are localized, x-ray therapy is effective in controlling the disease. As it disseminates, and the skin infiltrations become larger, a course of nitrogen mustard or TEM may cause a decrease in the size of the skin lesions and relief of pruritus. This improvement may sometimes last for 3 to 5 months, and if local lesions occur they can be treated by x-rays (49). In the far-advanced stages of the disease, HN2 has produced remarkable regressions of the disease for brief periods, sometimes for only 4 to 7 days, followed by rapid recurrence. Continued administration of HN2 will produce serious depression of the bone marrow without any further improvement in the disease Kennedy et al. (64) have described some degree of temporary improvement in 4 of 9 patients treated with urethane.

Polycythemia Vera

P^{32} appears to be the treatment of choice in polycythemia vera (115). Its use may relieve symptoms and keep the hematocrit within normal limits for long periods The occasional development of acute leukemia in polycythemia vera has been noted by many observers, and Schwartz and Ehrlich (99) have reviewed the evidence suggesting that P^{32} therapy may be leukemogenic in these patients Thus far, the data do not indicate that this is a common or necessarily direct result of P^{32} therapy.

HN2 is also effective, but the remissions induced last for 3 to 5 months, and are, thus, shorter than with P^{32} (108), furthermore, the nausea and vomiting induced by HN2 makes it less convenient to use. Oral TEM has overcome this objection, and it has been used successfully in several clinics

Multiple Myeloma

Multiple myeloma is a neoplastic disease of the plasma cells It is progressive, but there may be occasional periods of remission and exacerbation,

in the peripheral blood picture, and reticulum cell sarcoma. Transitional stages from one type of lymphosarcoma to another, and to the allied conditions of Hodgkin's disease and lymphatic leukemia, may occur in an individual patient. The natural course of this group of diseases varies remarkably, from tumors that have progressed with explosive dissemination and growth, to the indolent and relatively benign forms Sugarbaker and Craver reviewed the clinical course of 196 patients with lymphosarcoma, of whom 184 were diagnosed as reticulum cell sarcoma. The average survival of these patients was 18 6 months, and 15 9 per cent survived for 5 years or longer. The more favorable types of lymphosarcoma include: Brill-Symmers' disease (giant follicular lymphosarcoma) which is usually slowly progressive and responsive to x-ray therapy and nitrogen mustard and allied compounds for long periods; localized lymphosarcoma of the head and neck region, principally treated by x-rays and interstitial gold radon seeds, with a 52 per cent five-year survival rate (13), and primary reticulum cell sarcomas of the bone, where a 47.6 per cent five-year cure rate has been reported (16).

Apparently localized lymphosarcoma should be treated by surgical excision and/or intensive x-ray therapy. When the disease involves several areas, x-ray therapy is the most satisfactory method for controlling the process. In some of the patients with lymphosarcoma, particularly in the leukolymphosarcoma group, HN2 or TEM has induced prolonged regression of enlarged nodes and improvement in general condition. The best responses have occurred in patients with slowly progressive disease. These patients should be treated cautiously, since occasionally they may show an unusual sensitivity to the nitrogen mustard compounds, for example, the initial course of HN2 should total only 0.2 mg. per kg., and oral TEM begun at 5 mg (table 1), and treatment be continued and the dosage increased if the white cell and platelet counts do not fall, and the enlarged nodes have shown no signs of regression

In the aggressive forms or stages of the disease, HN2 and allied substances may produce some regression of the enlarged masses, but these responses are often distressingly brief In fact, following a course of HN2 or TEM, there may sometimes be an apparent acceleration in the growth of the tumor. This may be explained by the transient interruption in tumor growth produced by treatment, followed by a rapid recurrence While a debatable procedure, we have continued to give these patients a trial of HN2 or TEM, because of the occasional patient that has responded Furthermore, in patients with enlarged masses pressing on vital structures, a course of HN2 may cause considerable and rapid relief of symptoms, and x-ray therapy may be given more rapidly and effectively to the involved areas.

In the rapidly progressive stage, the disease grows so widely and rapidly

patients are cured by surgery under the best circumstances at present (26); about 95 per cent of the cases, thus, are inoperable or recur after surgery, and death usually occurs within one to 2 years. Palliative procedures in these patients are not very satisfactory and, at best, they prolong life for only short periods (75).

Radiotherapy is the most useful palliative measure (18). It can be directed to the involved areas, and it will produce regression of local disease, symptomatic improvement and, according to LaDue, an average prolongation of life of about 4 months Extrathoracic metastases may respond to x-ray therapy, and symptoms due to metastases in the brain, bone and liver have been relieved temporarily Occasionally a patient with circumscribed carcinoma of the lung has been cured by intensive radiotherapy, but this is not a consistent or predictable result and radiotherapy in lung cancer must be regarded as a palliative procedure.

Nitrogen mustard (HN2) has produced definite and consistent evidences of temporary improvement in lung cancer Karnofsky et al (61), in a series of 35 cases, analyzed the therapeutic responses as 1) immediate palliation and, 2) duration of improvement. Approximately 60 per cent of the cases obtained immediate partial or complete relief of cough, hemoptysis, dyspnea, weakness and pain, and in 8 of 9 patients with compression of the superior vena cava, there was a rapid lessening of symptoms and associated signs. Of the 35 patients treated, 17 (49 per cent) showed some evidence of objective improvement, 10 patients showed regression in pulmonary densities and increased aeration of the lung, 10 had a decrease in size of metastases, and in 2 pleural effusions decreased The duration of remission was brief, averaging about 3 weeks, but they were sometimes longer, particularly in the slower-growing tumors While the anaplastic carcinomas responded more dramatically and recurred rapidly, the more differentiated tumors were also affected temporarily Patients responding to the first course of nitrogen mustard obtained a lesser degree of improvement to the second course. Roswit and Kaplan (94, 95) reported 40 patients treated with HN2, 30 obtained remissions of severe systemic symptoms, and in 19 patients objective improvement occurred The remissions lasted an average of 3½ weeks (1 to 17 weeks) In patients with serious systemic symptoms, following the improvement produced by a course of HN2, it was possible to undertake a definitive course of x-ray therapy. These authors reviewed the reports in the literature of cases of lung cancers treated with HN2, of 254 treated, 134 (52 8 per cent) showed some type of favorable response While it is likely that HN2 may induce improvement by decreasing the inflammatory reaction in the tumor, Gaensler et al (38) have described cellular changes in lung cancers similar to those induced by x-ray therapy

Since TEM is believed to be similar to HN2 in its therapeutic activity,

and the average patient survives about 2 to 3 years from clinical onset. There are numerous abnormal findings associated fairly regularly with the disease, including bone marrow invasion, widespread bone destruction, elevated serum proteins and serum globulin, Bence-Jones proteinuria, splenic enlargement, anemia and renal involvement. These various manifestations in multiple myeloma have been summarized concisely by Limarzi (72).

X-ray therapy is the most effective method of controlling the local manifestations of multiple myeloma, by producing tumor regression in soft tissue and bone, and by relieving pain (40). Loge and Rundles (73) obtained striking improvement with urethane, with relief of pain, improvement in hemoglobin value and hematopoietic activity, reversal of the serum proteins toward normal and disappearance of Bence-Jones proteinuria (97) The most satisfactory responses were seen in the early stages of the disease In a group of 11 cases treated by Harrington and Moloney (48), 6 obtained clinical and laboratory evidence of improvement including relief of pain, gain in weight, increase in hemoglobin level, reduction in abnormal serum proteins, and suppression of myeloma cells in the marrow. They believe that the best responses occurred in chronic forms, and not in the acute blast form of the disease. Urethane should be given in adequate doses and continued usually for 2 to 4 weeks or more until clinical improvement results or toxicity, such as nausea and vomiting, or bone marrow depression intervenes. It is our impression, in reviewing patients in our clinic and in other series, that about 20 per cent may show some degree of improvement, and in an occasional patient it may be marked, there is, as yet, however, no evidence that the duration of life is appreciably prolonged

In the past 2 years ACTH and cortisone have been found to induce relief of pain, reduction in the serum proteins and a rise in hemoglobin level in occasional patients with multiple myeloma (83) There was no improvement in the bone marrow picture or in bone lesions by radiographic examination Usually the response lasts for a few weeks, but in some cases improvement has continued for almost one year The immature "blastic" forms of multiple myeloma appear to be most responsive to the adrenocortical hormones (77).

Nitrogen mustard has relieved pain and produced regression of palpable masses, but it has had no important influence on the disease (54) The folic acid antagonists have been of no value, and stilbamidine has had no regular action in the relief of pain. Lawrence and Wasserman (70) treated 20 patients with radioactive phosphorus, and observed some measure of clinical improvement in 25 per cent of the patients

Lung Cancer

The lung is one if the most common sites of cancer and lung cancer appears to be increasing in frequency Only about 5 to 8 per cent of the

Breast Cancer

As a result of early diagnosis, radical mastectomy and, in some cases, local x-ray therapy, about 40 per cent of all patients with breast cancer survive for 5 years or longer without evidence of disease The remainder of the patients (this is a large group composed of postoperative recurrences or patients who are inoperable when first seen) will require palliative therapy. Breast cancer runs a variable and sometimes slowly progressive course, so that it is difficult to evaluate the effectiveness of various palliative procedures Treatment must be individualized for each patient, and the following considerations may define the indications for hormonal therapy in inoperable breast cancer.

In patients in whom symptoms are produced by well-defined lesions, localized x-ray therapy may produce marked and prolonged relief, without appreciable systemic reactions. Garland et al. (39), in reviewing the response of osseous metastases of breast cancer to x-rays, observed relief of pain in 70 per cent of the patients for periods ranging from 50 to 100 per cent of their survival time in 75 per cent of the responsive cases Radiation therapy to local recurrences, pulmonary and liver metastases may also be of temporary benefit.

Castration may also be an effective palliative procedure in the pre-menopausal patient and in early post-menopausal patients. Castration may be accomplished surgically or by irradiation of the ovaries; the former is more certain, the response more rapid and the method to be preferred. Nathanson and Kelley have noted a trend toward surgical castration in young women, and x-ray castration in women over 40 where ovarian activity is beginning to wane.

Androgen therapy is indicated in the pre-menopausal and up to 10 years post-menopausal patient with wide-spread disease who has not responded to castration Symptomatic improvement is striking in about 70 per cent of the patients with relief of pain, weakness, dyspnea and anorexia In some cases, despite symptomatic improvement, the disease continues to progress. Twenty to 25 per cent of the patients showed objective evidence of benefit, with some recalcification of osteolytic lesions, healing of pathological fractures and an apparent interruption in the formation of new lesions Some patients also showed a decrease in size of metastatic nodes, pulmonary infiltrations and enlarged livers In some instances, lesions have regressed while others progressed

While symptomatic relief occurs within 2 to 4 weeks, objective improvement may be more delayed Treatment, consequently, should be continued for at least 3 months if the patient's condition is not becoming worse, before deciding that testosterone is without benefit. The average duration of improvement is about 6 months, although some patients have been

it is to be expected that oral and intravenous TEM will have some effect in lung cancer. In our brief experience, however, we have observed only one favorable response to oral TEM in 5 cases treated; this series is inconclusive, and further data are necessary.

In recurrent pleural effusions, intrapleural HN2 may sometimes decrease or control the formation of fluid. Our procedure consists in removing almost all the pleural effusion, and then 10 to 20 mg. of HN2 is injected into free fluid in the pleural space; the total dose given is in the range of 0 4 to 0 6 mg. per kg. In some patients this procedure appears to induce an obliterative pleuritis, in others the HN2 is absorbed and causes the same systemic effects seen from the intravenous injection of HN2.

Indications for the use of HN2 in lung cancer may be summarized as follows:

FOR RELIEF OF THE SUPERIOR VENA CAVA SYNDROME

Patients with severe symptoms due to compression of the superior vena cava by tumor may obtain marked relief from a single intravenous dose of 0 4 mg. per kg Because of the elevated venous pressure in the arm, it is well to give the injection with the arm elevated In some cases following HN2 injection diffuse thrombosis has resulted in the superficial veins of the arm and shoulder, but we have not seen pulmonary emboli from the thrombosed area. Following relief of the syndrome within 2 to 3 days, it is often advisable to begin x-ray therapy to the involved area in the hope of controlling the disease for longer periods Usually, however, metastases subsequently appear in other areas, and effective control of the primary lesion does not necessarily prolong life In some cases repeated courses of HN2, as permitted by the peripheral blood count, may palliate the disease as effectively as x-ray therapy.

FOR RAPID RELIEF OF SYMPTOMS IN EXTENSIVE LUNG CANCER, TO BE FOLLOWED BY LOCALIZED IRRADIATION

Intensive radiotherapy to the chest has not aggravated the leukopenia induced by a course of HN2, and it seems reasonable to give the entire course of palliative radiotherapy within a short period, within 8 to 10 days, in some cases, following HN2, rather than to prolong treatment by greater fractionation of the dose

IN PATIENTS TREATED BY X-RAY THERAPY AND NO LONGER RESPONDING TO X-RAYS

Occasionally such patients may show some improvement and the symptoms from extra-thoracic metastases may be relieved

prolongation of life. Nesbit and Baum (78), in an analysis of 1818 patients, presented data (shown in table 7) on the effects of castration, estrogen and combined castration and estrogen therapy in prostatic cancer.

About 70 per cent of the patients with prostatic cancer show an early response to estrogen therapy or castration but, as noted in the above data, combined castration and estrogen therapy gave the longest survival in the patients without metastases at their first admission, and in the patients with metastases when first seen, castration or combined estrogen and castration gave the best response. Patients treated initially with estrogens who had relapsed obtained some improvement after castration, the reverse procedure rarely was successful. Nesbit and Baum's data appear to demonstrate convincingly that hormonal treatment will definitely prolong life in prostatic cancer, most strikingly in patients who have no metastases when first seen.

TABLE 7

	NO METASTASES AT FIRST ADMISSION		METASTASES AT FIRST ADMISSION	
	% Dead in 3 yrs	% Dead in 5 yrs	% Dead in 3 yrs	% Dead in 5 yrs
Untreated	78	90	89	94
Estrogen	50 3	71	74	90
Castration	46 6	69	63 6	79
Estrogen & castration	34	56	64 8	80

The most effective method of managing the patient with inoperable prostatic cancer is still a matter of continuing debate (77). If urinary obstruction is present and does not respond promptly to castration, it should be relieved by surgery The present trend in hormonal therapy appears to be the initiation of treatment as soon as the diagnosis is established, rather than waiting, as some recommend, until definite symptoms develop Orchiectomy gives the most rapid therapeutic response, within a matter of one to 2 days, whereas the response to estrogen therapy is slower, requiring one to 2 weeks, in general, however, the results of both treatments are similar. Castration followed immediately by estrogen therapy is believed by many writers to be the most effective method of treatment.

Following treatment and depending on the clinical situation, the responsive patient may show a rapid relief of pain, decrease in urinary tract obstruction, diminution in the size of an enlarged prostate, shrinkage of metastatic nodes and soft tissue masses, and a reduction in an elevated serum acid phosphatase level The patient regains weight, his general condition improves, and the condition of a seriously sick patient may be remarkably alleviated so that he is able to return to his normal activities

In most cases, after intervals of one to 3 years, the disease begins to

benefitted for much longer periods, and there is some evidence, as stated by Nathanson and Kelley (77), that the mean survival of patients responding to androgen therapy is about twice that of the unresponsive cases.

In some patients who improved and then relapsed while on androgen therapy, cessation of treatment has resulted in a second period of improvement. In 11 such patients treated by Escher (58) 7 exhibited a secondary period of improvement beginning one to 4 months after cessation of testosterone and lasting for 2 to 19 months. It is suggested that some of the tumor cells actually become dependent on androgenic stimulation, and when this is withdrawn they temporarily regress. Estrogen therapy is not recommended in the pre-menopausal patient because, in some cases, it appeared to accelerate the course of the disease. In patients completely unresponsive to testosterone, however, a trial of estrogens is worthwhile. Adrenalectomy appears to induce temporary improvement in some of these far-advanced cases, and it is now under investigation (51).

Estrogens are used in the patients 10 years post-menopausal or over 60 years of age. These patients may show regression of soft tissue tumors, pulmonary infiltration and healing of skin ulcers in 40 to 50 per cent of the cases, and about 20 per cent of the osseous lesions respond, subjective improvement, such as relief of pain, dyspnea, anorexia and malaise, was observed in about 60 per cent of the patients. Improvement may be evident within 2 to 4 weeks, or may not appear until several months after continuous therapy, so estrogens should be given for at least 6 months, if the patient is not becoming worse on treatment. Improvement usually lasts for 6 months and frequently for much longer periods. Treatment should be continued during the period of improvement. Androgens may be more effective than estrogens in the treatment of osseous lesions in all age groups.

Breast cancer in males may show a striking response to castration, with regression of soft tissue and osseous metastases (120)

The serious side-effects of hormonal therapy, other than the virilizing and feminizing actions, should be noted again; these are fluid retention which may lead to cardiac failure, and hypercalcemia, particularly in the patients with osseous involvement These effects should be watched for carefully, and treated if they develop

Prostatic Cancer

Carcinoma of the prostate occurs most commonly in the older age group, it runs a very variable course, and some patients may be asymptomatic without treatment for many years At present only about 5 per cent of the patients can be cured by surgery, the remaining patients have recurrent, inoperable or metastatic disease which requires palliative therapy By the use of castration and/or estrogen therapy, there has been a marked change in the clinical manifestations of the disease; and in some cases a definite

prolongation of life. Nesbit and Baum (78), in an analysis of 1818 patients, presented data (shown in table 7) on the effects of castration, estrogen and combined castration and estrogen therapy in prostatic cancer.

About 70 per cent of the patients with prostatic cancer show an early response to estrogen therapy or castration but, as noted in the above data, combined castration and estrogen therapy gave the longest survival in the patients without metastases at their first admission, and in the patients with metastases when first seen, castration or combined estrogen and castration gave the best response. Patients treated initially with estrogens who had relapsed obtained some improvement after castration; the reverse procedure rarely was successful. Nesbit and Baum's data appear to demonstrate convincingly that hormonal treatment will definitely prolong life in prostatic cancer, most strikingly in patients who have no metastases when first seen.

TABLE 7

	NO METASTASES AT FIRST ADMISSION		METASTASES AT FIRST ADMISSION	
	% Dead in 3 yrs	% Dead in 5 yrs	% Dead in 3 yrs	% Dead in 5 yrs
Untreated	78	90	89	94
Estrogen .	50.3	71	74	90
Castration	46 6	69	63 6	79
Estrogen & castration	34	56	64 8	80

The most effective method of managing the patient with inoperable prostatic cancer is still a matter of continuing debate (77). If urinary obstruction is present and does not respond promptly to castration, it should be relieved by surgery. The present trend in hormonal therapy appears to be the initiation of treatment as soon as the diagnosis is established, rather than waiting, as some recommend, until definite symptoms develop. Orchiectomy gives the most rapid therapeutic response, within a matter of one to 2 days, whereas the response to estrogen therapy is slower, requiring one to 2 weeks, in general, however, the results of both treatments are similar. Castration followed immediately by estrogen therapy is believed by many writers to be the most effective method of treatment

Following treatment and depending on the clinical situation, the responsive patient may show a rapid relief of pain, decrease in urinary tract obstruction, diminution in the size of an enlarged prostate, shrinkage of metastatic nodes and soft tissue masses, and a reduction in an elevated serum acid phosphatase level. The patient regains weight, his general condition improves, and the condition of a seriously sick patient may be remarkably alleviated so that he is able to return to his normal activities.

In most cases, after intervals of one to 3 years, the disease begins to

extend. Increase in estrogen dosage, or castration in a patient who has been maintained on estrogen therapy, may give another period of improvement, but the disease gradually becomes resistant to hormonal control therapy. The use of nitrogen mustard, x-ray therapy, urethane and progesterone has occasionally given temporary relief to some of these patients At present the effect of adrenalectomy in these resistant patients is being studied and there is already evidence that some cases may show a temporary response.

Thyroid Cancer

Radioactive iodine is destructive to thyroid carcinoma if it can be selectively concentrated in the tumor In a study of 100 thyroid carcinomas by Fitzgerald and Foote (35), 46 per cent showed some uptake of radioactive iodine, and these were principally of the solid, alveolar and follicular types. The concentration of I^{131} in many instances was slight, and it is not distributed homogeneously throughout the tumor. By removal of the normal thyroid gland, the prolonged administration of large doses of thiouracil, a significant increase in the uptake of I^{131} by some metastases can be obtained. If tracer doses of I^{131} demonstrate that the metastases will concentrate I^{131} to a satisfactory degree, a calculated therapeutic dose of I^{131}, in the range of 100 to 500 millicuries, is given

Seidlin et al (101) studied 30 patients with metastatic thyroid carcinoma of whom 12 were treated with I^{131}, in 8 some degree of improvement was obtained In the series of 146 patients studied by Rawson et al (89), in 20 it was "desirable and possible to administer therapeutic amounts of radioactive iodine", and in 10 objective evidence of tumor destruction was observed. In these selected patients, striking responses have sometimes occurred, with regression of pulmonary and osseous metastases, healing of bone lesions and improvement in general condition. The relatively low incidence of thyroid cancer and the variability of its course make it difficult to assess the contribution that I^{131} has made to the control of this disease. In the few thyroid cancers which can concentrate an adequate amount of I^{131}, a therapeutic trial of this agent is warranted

Miscellaneous Tumors

There has been no consistent or predictable clinically useful response of the tumors already summarized to other chemotherapeutic agents, or of other types of tumors to the chemotherapeutic agents presented here There have been isolated reports of remarkable improvement following certain agents, which may or may not be directly related to the treatment. For example, Lemon et al (71) reported a remission in osseous metastases from adenocarcinoma of the thyroid associated with testosterone propionate injections; Freed et al. (37) observed regression of pulmonary metastases

from adenocarcinoma of the body of the uterus on androgens; Bayrd and Hall (3) reported a prolonged remission in plasma cell leukemia with P^{32}, and Goodman and Lewis (44) obtained regression of metastatic carcinoma to the skin with urethane. We have observed with HN2 a remarkable 8-month remission in a chorioepithelioma, and Farber (32) has presented a patient whose metastatic melanoma disappeared following TEPA (table 2)

Supportive measures are extremely important and they include an adequate diet, proper management of sedation, encouragement of the patient and careful attention to new symptoms as they appear. Occasional blood transfusions, particularly if the hemoglobin level is falling, antibiotics for evidences of intercurrent infection, testosterone and, in some cases, a trial of cortisone or ACTH (although there has been no objective evidence that these agents inhibit the growth of carcinoma) may contribute to the patient's general well-being.

Tumors in Children

Acute leukemia, which comprises about one-half of neoplastic disease in children, has already been discussed. Thyroid cancer in children has been reviewed by Duffy and Fitzgerald (29); the disease runs a variable course, and an occasional patient may be suitable for I^{131} therapy The most important tumors in children, excluding brain tumors, are Wilm's tumor, neuroblastoma and osteogenic sarcoma. Widespread neuroblastomas may show partial and temporary regression from nitrogen mustard and its derivatives, or from a course of the folic acid antagonists. Radiation therapy is used concurrently, when indicated, against circumscribed lesions Inoperable or recurrent Wilm's tumors are also best controlled by x-ray therapy, but, in extensive disease, HN2 or related substances may induce temporary regression. In general, children's tumors are more sensitive than adult tumors to chemotherapeutic agents, and a course of nitrogen mustard may induce a prompt regression in a bulky tumor with relief of symptoms. Eosinophilic granulomas and related reticuloendothelioses may show a very striking response to x-ray therapy (14). Prolonged clinical remissions in widespread disease have been obtained with HN2 and with the folic acid antagonists (24).

Regional Chemotherapy

Klopp et al (66) and Bierman et al (4, 6) demonstrated the feasibility of injecting nitrogen mustard into the blood supply of tumor-bearing areas, and they have reported temporary regressions and histological changes in the tumor Klopp's method consists in placing a plastic catheter into the artery to the involved area, injecting HN2 daily in small doses (1 to 2 mg. three times daily) and the treatment may be continued for one to 3 weeks.

Severe bone marrow depression is the limiting factor in treatment (2) and usually about 50 *per cent more HN2* is tolerated by this route than by the intravenous route. Tumors of the head and neck and in the pelvis (20) have been treated by this method. Bierman et al. have developed a method for catheterizing the desired artery, and carcinomas in the liver and tumors of the extremities have been treated by injecting the hepatic and femoral arteries, respectively, with single large doses of HN2. There may be immediate relief of local symptoms and decrease in tumor size; but these remissions appear to be of short duration. The role of regional chemotherapy with HN2, and the potentialities of this technique, in the treatment of localized inoperable cancer are now under investigation.

CONCLUSION

The management of inoperable neoplastic disease may be improved in many situations by the use of specific chemotherapeutic agents. These drugs should be used on the basis of specific indications, with knowledge of their mechanism of action and the hazards associated with their administration. The correct application of these agents implies that the physician is aware of the exact diagnosis and extent of the patient's disease, the natural history of his type of cancer and the effectiveness of other forms of treatment. Chemotherapy should be considered only after the possibility of a curative procedure has been ruled out, and in these situations it has gained a place, along with radiation therapy and surgery, in the palliative treatment of cancer

BIBLIOGRAPHY

1. ANGIER, R B, BOOTHE, J H., HUTCHINGS, B L, MOWAT, J H, SEMB, J, STOKSTAD, E L R, SUBBAROW, Y, WALLER, C W, COSULICH, D. B, FAHRENBACH, M J, HULTQUIST, M E, KUH, E, NORTHEY, E H, SEEGER, D R, SICKELS, J P AND SMITH, J M, JR · The structure and synthesis of the liver L casei factor Science, 103: 667, 1946
2. BATEMAN, J C, KLOPP, C T AND CROMER, J K · Hematologic effects of regional nitrogen mustard therapy Blood, 6: 26, 1951
3. BAYRD, E D AND HALL, B E Unusual remission after radiophosphorus therapy in a case of "acute plasma cell leukemia" Blood, 3. 1019, 1948
4. BIERMAN, H R, BYRON, R L, JR AND KELLY, K H. Therapy of inoperable visceral and regional metastases by intra-arterial catherization in man (abst) Cancer Res, 11: 236, 1951
5. BIERMAN, H. R, COHEN, P, McCELLAND, J N AND SHIMKIN, M B Effect of [illegible] life in children with lympho- [illegible]
6. [illegible] JR, DOD, K S., KELLY, K H [illegible] tion of viscera in man Am J Roentgenol, 66: 555, 1951
7. BUCKLEY, S. M, STOCK, C C, PARKER, R. P, CROSSLEY, M. L., KUTT, E AND

SEEGER, D R.: Inhibition studies of some phosphoramides against sarcoma 180 Proc Soc. Exper Biol & Med, 78: 209, 1951.
8. BURCHENAL, J H. The newer nitrogen mustard in the treatment of leukemia. Radiology, 50: 494, 1948.
9. BURCHENAL, J. H, GOETCHIUS, S. K., STOCK, C. C. AND HITCHINGS, G H: Diamino dichlorophenyl pyrimidines in mouse leukemia (abst.). Cancer Res, 12: 251, 1952.
10. BURCHENAL, J. H, KARNOFSKY, D A., KINGSLEY-PILLERS, E. M, SOUTHAM, C. M, MYERS, W. P L, ESCHER, G. C, CRAVER, L F., DARGEON, H. W AND RHOADS, C P.: The effects of the folic acid antagonists and 2,6-diamino purine on neoplastic disease Cancer, 4: 549, 1951
11. BURCHENAL, J. H., LESTER, R A, RILEY, J. B AND RHOADS, C P: Studies on the chemotherapy of leukemia I Effect of certain nitrogen mustards and carbamates on transmitted mouse leukemia Cancer, 1: 339, 1948
12. BURCHENAL, J. H, MYERS, W. P. L, CRAVER, L. F. AND KARNOFSKY, D A. The nitrogen mustards in the treatment of leukemia Cancer, 2: 1, 1949
13. CATLIN, D S Lymphosarcoma of the head and neck Am J. Roentgenol, 59: 354, 1948
14. CHILDS, D S, JR AND KENNEDY, R L J.: Reticulo-endotheliosis of children treatment with roentgen rays Radiology, 57: 653, 1951
15. CLARKE, D. A, BUCKLEY, S M., STERNBERG, S S, STOCK, C C, RHOADS, C P. AND HITCHINGS, G H. Effects of 2,4-diaminopyrimidines on mouse sarcoma 180 (abst). Cancer Res., 12: 255, 1952
16 COLEY, B L, HIGINBOTHAM, N L. AND GROESBECK, H P. Primary reticulum-cell sarcoma of bone Radiology, 55: 641, 1950
17 Council on Pharmacy and Chemistry, Subcommittee on Steroids and Cancer. Current status of hormone therapy in advanced mammary cancer J A M A, 146: 471, 1951
18 CRAVER, L F Bronchogenic carcinoma Am. J Roentgenol, 43: 469, 1940
19 CRAVER, L F Hodgkin's disease: Tice's Practice of Medicine, 5: 107, W F Prior Co, Inc, Maryland, 1951
20 CROMER, J K, BATEMAN, J C, BERRY, G N, KENNELLY, J. M, KLOPP, C T. AND PLATT, L I Use of intra-arterial nitrogen mustard therapy in the treatment of cervical and vaginal cancer. Am J Obs & Gyn, 63: 538, 1952
21 CRUZ, W O. AND MOUSSATACHE, H. Acute thrombocytopenia induced in dogs by administration of urethane (ethyl carbamate) Blood, 3: 793, 1948
22 CUSTER, R P An Atlas of Blood and Bone Marrow Saunders and Co, Phila, 1949
23 DAMESHEK, W, WEISFUSE, L AND STEIN, T Nitrogen mustard therapy in Hodgkin's disease Blood, 4: 338, 1949
24. DARGEON, H W Personal communication
25 DARTE, J M M, SNELLING, C E, LASKE, B, JACKSON, S H AND DONOHUE, W L ACTH and cortisone in the treatment of leukemia in children. Canad Med Assoc J, 65: 566, 1951
26 DEBAKEY, M E, OCHSNER, A AND DECAMP, P Carcinoma of the lung. Monographs on Surgery, B N Carter (Edit), p 77, The Williams & Wilkins Co, 1950
27 DIAMOND, H D AND CRAVER, L F Radioactive phosphorus II. In the treatment of myeloid leukemia Cancer, 4: 990, 1951.
28 DIAMOND, H D, CRAVER, L F, WOODARD, H Q AND PARKS, G H.: Radio-

Severe bone marrow depression is the limiting factor in treatment (2) and usually about 50 per cent more HN2 is tolerated by this route than by the intravenous route. Tumors of the head and neck and in the pelvis (20) have been treated by this method. Bierman et al. have developed a method for catheterizing the desired artery, and carcinomas in the liver and tumors of the extremities have been treated by injecting the hepatic and femoral arteries, respectively, with single large doses of HN2. There may be immediate relief of local symptoms and decrease in tumor size; but these remissions appear to be of short duration The role of regional chemotherapy with HN2, and the potentialities of this technique, in the treatment of localized inoperable cancer are now under investigation.

CONCLUSION

The management of inoperable neoplastic disease may be improved in many situations by the use of specific chemotherapeutic agents. These drugs should be used on the basis of specific indications, with knowledge of their mechanism of action and the hazards associated with their administration. The correct application of these agents implies that the physician is aware of the exact diagnosis and extent of the patient's disease, the natural history of his type of cancer and the effectiveness of other forms of treatment. Chemotherapy should be considered only after the possibility of a curative procedure has been ruled out; and in these situations it has gained a place, along with radiation therapy and surgery, in the palliative treatment of cancer

BIBLIOGRAPHY

1 Angier, R B, Boothe, J H, Hutchings, B L, Mowat, J H, Semb, J, Stokstad, E L R, SubbaRow, Y, Waller, C. W, Cosulich, D B, Fahrenbach, M J, Hultquist, M E, Kuh, E, Northey, E H, Seeger, D R, Sickels, J P and Smith, J M, Jr The structure and synthesis of the liver L casei factor Science, 103: 667, 1946

2. Bateman, J C, Klopp, C T and Cromer, J K Hematologic effects [illegible] therapy Blood 6: 26 1951

[illegible]

4 Bierman, H R, Byron, R L, Jr and Kelly, K H Therapy of inoperable visceral and regional metastases by intra-arterial catherization in man (abst) Cancer Res, 11: 236, 1951

5 Bierman, H R, Cohen, P, McCelland, J N and Shimkin, M B Effect of [illegible] life in children with lympho- [illegible]

6 [illegible] Jr, Dod, K S, Kelly, K H and Black, [illegible] [illegible]tion of viscera in man Am J Roentgenol, 66: 555, 1951

7. Buckley, S M, Stock, C C, Parker, R. P, Crossley, M L, Kutt, E and

48 HARRINGTON, W. J AND MOLONEY, W C. The treatment of multiple myeloma with urethane Cancer, 3: 253, 1950
49 HENSTELL, H. H , TOBER, J N. AND NEWMAN, B A.: The influence of nitrogen mustard on mycosis fungoides Blood, 2: 564, 1947.
50 HIRSCHBOECK, J. S , LINDERT, M. C. F., CHASE, J AND CALVY, T. L : Effects of urethane in the treatment of leukemia and metastatic malignant tumors J A. M A , 136: 90, 1948
51. HUGGINS, C AND BERGENTHAL, D M . Surgery of the adrenals J A .M A , 147: 101, 1951.
52 INGLE, D J.: The biologic properties of cortisone: a review J Clin Endocrinol 10: 1312, 1950
53 KARNOFSKY, D. A.: Medical progress chemotherapy of neoplastic disease. New Eng. J Med , 239: 226, 260, 299, 1948
54 KARNOFSKY, D A Nitrogen mustards in the treatment of neoplastic disease. Advances in Int. Med , 4: 1, Year Book Publishers, Inc , Chicago, 1950
55 KARNOFSKY, D A , ABELMANN, W , CRAVER, L F AND BURCHENAL, J. H The use of nitrogen mustards in the palliative treatment of bronchogenic carcinoma Cancer, 1: 634, 1948
56 KARNOFSKY, D A AND BURCHENAL, J. H . Present status of clinical cancer chemotherapy Am J Med , 8: 767, 1950
57. KARNOFSKY, D A , BURCHENAL, J H , ARMISTEAD, G C , JR , SOUTHAM, C M , BERNSTEIN, J. L , CRAVER, L. F AND RHOADS, C P. Triethylene melamine in the treatment of neoplastic disease Arch Int Med , 87: 477, 1951.
58 KARNOFSKY, D A , BURCHENAL, J H AND ESCHER, G C Chemotherapy of neoplastic diseases Medical Clinics of North America (Nationwide Number), p 439, W B Saunders Company, Philadelphia, 1950
59 KARNOFSKY, D A , BURCHENAL, J H , ORMSBEE, R A , CORNMAN, I. AND RHOADS, C P . Experimental observations on the use of nitrogen mustards in the treatment of neoplastic diseases Approaches to Tumor Chemotherapy, p 293, Washington, D C , Am Assoc Advancement Sci , 1947
60 KARNOFSKY, D A , CRAVER, L F , RHOADS, C P. AND ABELS, J C An evaluation of methyl bis-(B chloroethyl) amine hydrochloride and tris-(B-chloroethyl) amine hydrochloride (nitrogen mustards) in the treatment of lymphomas, leukemia and allied diseases Approaches to Tumor Chemotherapy, p 319, Washington, D C , Am Assoc Advancement Sci , 1947
61. KARNOFSKY, D A , GRAEF, I. AND SMITH, H W. Studies on the mechanism of action of the nitrogen and sulfur mustards *in vivo* Am J Path , 24: 275, 1948
62 KARNOFSKY, D A , PATTERSON, P A AND RIDGWAY, L P Effect of folic acid, "4 amino" folic acids and related substances on growth of chick embryo Proc Soc Exper Biol & Med , 71: 447, 1949
63 KASDON, S C , FISHMAN, W H , DART, R M , BONNER, C D AND HOMBURGER, F Methylandrostenediol in palliative treatment of breast cancer. J A M A , 148: 1212, 1952
64 KENNEDY, B J , NATHANSON, I T AND AUB, J C Ethyl carbamate (urethane) in the treatment of mycosis fungoides Cancer, 3: 66, 1950
65 KINGSLEY-PILLERS, E M , BURCHENAL, J H , ELIEL, L P AND PEARSON, O H Studies on the development of resistance to chemotherapeutic agents—A-methopterin, cortisone, and ACTH in children with acute leukemia J A M A , 148: 987, 1952
66 KLOPP, C T , ALFORD, T C , BATEMAN, J , BERRY, G N. AND WINSHIP, T :

active phosphorus I. In the treatment of lymphatic leukemia Cancer, 3: 779, 1950

29. Duffy, B. J., Jr and Fitzgerald, P. S Cancer of the thyroid in children; a report of 28 cases. J Clin Endocrinol, 10: 1296, 1950.

30. Dustin, P · Cytological action of ethylcarbamate (urethane) and other carbamic esters in normal and leukaemic mice and in rabbits. Brit. J. Cancer, 1: 48, 1947.

31. Dyer, H M : An index of tumor chemotherapy, p. 320 National Cancer Institute, National Institute of Health, Washington, D. C, 1949.

32 Farber, S. (Edit.) Proceedings of the second conference on folic acid antagonists in the treatment of leukemia Blood, 7: (Suppl): 97, 1952

33. Farber, S, Diamond, L K, Mercer, R. D, Sylvester, R F, Jr and Wolff, J A Temporary remissions in acute leukemia in children produced by folic acid antagonist, 4-aminopteroyl-glutamic acid (aminopterin) New Eng J Med, 238: 787, 1948

34. Farber, S. and Downing, V Lymphoma and leukemia panel. Second National Cancer Conference, 1952

35 Fitzgerald, P. J and Foote, F W, Jr The function of various types of thyroid carcinoma as revealed by the radioautographic demonstration of radioactive iodine (I^{131}) J Clin Endocrinol, 9: 1153, 1949

36 Forkner, C E Leukemia and Allied Diseases New York, MacMillan Co, 1938

37 Freed, J H, Pendergrass, E P and Carnwath, J W · Androgen therapy in control of pulmonary metastases from adenocarcinoma of corpus uteri Am J Roentgenol, 65: 596, 1951

38 Gaensler, E A, McKay, D G, Ware, P F and Lynch, J P Cytological changes in bronchogenic carcinoma following treatment with nitrogen mustard (methyl-bis-(B-chloroethyl) amine) Arch Path, 46: 503, 1948

39 Garland, L H, Baker, M, Picard, W H, Jr and Sisson, M A Roentgen and steroid hormone therapy in mammary cancer metastatic to bone J A M A, 144: 997, 1950

40 Garland, L H and Kennedy, B R Roentgen treatment of multiple myeloma Radiology, 50: 297, 1948

41 Gellhorn, A and Collins, V P A quantitative evaluation of the contribution of nitrogen mustard to the therapeutic management of Hodgkin's disease Ann Int Med, 35: 1250, 1951

42 Gellhorn, A and Jones, L D Chemotherapy of malignant disease Am J Med, 6: 188, 1949

43 Goldsmith, E D and Harnly, M H Reversal of the action of a folic acid antagonist, 4 aminopteroylglutamic acid, in Drosophila melanogaster (abstr) Cancer Res, 10: 220, 1950

44. Goodman, M J and Lewis, H. P Urethane in leukemia J A M A, 132: 1105, 1946

45 Goodman, L S, Wintrobe, M M, Dameshek, W, Goodman, M J, Gilman, A and McLennan, M T Nitrogen mustard therapy, use of methyl-bis (B-chloroethyl) amine hydrochloride and tris-(B-chloroethyl) amine hydrochloride for Hodgkin's disease, lymphosarcoma, leukemia and certain allied

of cortisone on production of granulation tissue in the rabbit. Proc. Soc. Exp Biol. & Med., 72: 718, 1949.

89. RAWSON, R. W, RALL, J. E. AND PEACOCK, W. Limitations and indications in the treatment of cancer of the thyroid with radioactive iodine J Clin. Endo, 11: 1128, 1951.

90. REINHARD, E. H, GOOD, J. T. AND MARTIN, E · Chemotherapy of malignant neoplastic diseases. J. A. M. A, 142: 383, 1950

91. RHOADS, C. P: Nitrogen mustards in the treatment of neoplastic disease. J A. M A, 131: 656, 1946.

92 RODGERS, C. L., DONOHUE, W. L. AND SNELLING, C E.· Leukaemia in children. Canad Med. Assoc. J., 65: 548, 1951.

93 ROSENTHAL, M. C, SAUNDERS, R H, SCHWARTZ, L. J, ZANNOS, L, SANTIAGO, E P. AND DAMESHEK, W.: The use of adrenocorticotropic hormone and cortisone in the treatment of leukemia and leukosarcoma. Blood, 6: 804, 1951

94. ROSWIT, B. AND KAPLAN, G.· Role of nitrogen mustard (HN_2) as a systemic adjunct to the radiation therapy of certain malignant diseases. Am J. Roentgenol, 61: 626, 1949.

95 ROSWIT, B AND KAPLAN, G. Nitrogen mustard as an adjunct to radiation in the management of bronchogenic cancer Radiology, 57: 394, 1951

96 RUNDLES, R. W. AND BARTEN, W. B Triethylene melamine in the treatment of neoplastic disease Blood, 7: 483, 1952

97 RUNDLES, R W, DILLON, M. L. AND DILLON, E S Multiple myeloma III Effect of urethane therapy on plasma cell growth, abnormal serum protein components and Bence-Jones proteinuria J Clin Invest, 29: 1243, 1950.

98 SCHULMAN, I, LANMAN, J R, LAXDAHL, D E AND HOLT, L E., JR · Effects of ACTH and cortisone on leukemia in childhood Pediatrics, 8: 34, 1951.

99. SCHWARTZ, S. O AND EHRLICH, L The relationship of polycythemia vera to leukemia; a critical review Acta Hemato, 4. 129, 1950

100 SEGALOFF, A, GORDON, D, HORWITT, B N, SCHLOSSER, J V AND MURISON, P J.. Hormonal therapy in cancer of the breast. II. Effect of methylandrostenediol on clinical course and hormonal excretion Cancer, 5: 271, 1952

101 SEIDLIN, S. M, ROSSMAN, I., OSHRY, E AND SIEGEL, E Radioiodine therapy of metastases from carcinoma of the thyroid a six-year progress report J Clin Endo, 9: 1122, 1949

102 SHIMKIN, M B AND BIERMAN, H R Experimental chemotherapy of neoplastic diseases Radiology, 53: 518, 1948

103 SHIMKIN, M B, METTIER, S R AND BIERMAN, H R Myelocytic leukemia an analysis of incidence, distribution, and fatality, 1910–1948 Ann. Int Med, 35: 194, 1951.

104 SILVERBERG, G. M AND DAMESHEK, W Use of triethylene melamine in treatment of leukemia and leukosarcoma J A M A, 148· 1015, 1952

105 SKIPPER, H E AND BRYAN, C E Carbamates in the chemotherapy of leukemia. III The relationship between chemical structure and antileukemic action of a series of urethan derivatives J Nat Cancer Inst, 9: 391, 1949.

106 SMITH, T R., JACOBSON, L O, SPURR, C L, ALLEN, J G AND BLOCK, M H A coagulation defect produced by nitrogen mustard Science, 107: 474, 1948

107 SOUTHAM, C M, CRAVER, L F, DARGEON, H W AND BURCHENAL, J H A study of the natural history of acute leukemia with special reference to the duration of the disease and the occurrence of remission Cancer, 4: 39, 1951

108 SPURR, C L., SMITH, T R, BLOCK, M AND JACOBSON, L O A clinical study

Fractionated intra-arterial cancer chemotherapy with methyl-bis-amine hydrochloride, a preliminary report. Ann. Surg , **132:** 811, 1950
67. Kravitz, S. C , Diamond, H D. and Craver, L F · Uremia complicating leukemia chemotherapy; report of a case treated with triethylene melamine J. A M. A , **146:** 1595, 1951.
68. LaDue, J. S.. Personal communication.
69. Law, L. W.: Urethane (ethyl carbamate) therapy in spontaneous leukemias in mice Proc Nat Acad Sci., **33:** 204, 1947.
70 Lawrence, J. H. and Wasserman, L. R · Multiple myeloma· a study of 24 patients treated with radioactive isotopes (P^{32} and Sr^{89}). Ann. Int. Med , **33:** 41, 1950
71. Lemon, H. M , Ravin, I S , Ross, J F., Sisson, J. H., Anglem, T. J and Branca, A W : *Testosterone therapy of metastatic adenocarcinoma of the thyroid*, with remission; case report. Cancer, **4:** 1176, 1951.
72 Limarzi, L. R : Diagnostic and therapeutic aspects of multiple myeloma Med Clin N Am , **35:** 189, 1951. W. B Saunders Co , Phila.
73. Loge, J P and Rundles, R. W. Urethane (ethyl carbamate) therapy in multiple myeloma Blood, **4:** 201, 1949.
74 Matthews, W B . A trial of B-naphthyldi-2-chloroethylamine (R48) in leukaemia, Hodgkin's disease, and allied diseases Lancet, **1:** 896, 1950
75 Mayer, E. and Roswit, B Newer palliative measures in the management of inoperable bronchogenic carcinoma Dis Chest, **21:** 491, 1952.
76 Moeschlin, S and Bodmer, A · Urethane-caused blood and bone marrow changes in agranulocytosis and panmyelopathy of the cat Blood, **6:** 242, 1951
77 Nathanson, I T and Kelley, R M Medical progress hormone treatment of cancer New Eng J Med , **246:** 135, 180, 1952
78 Nesbit, R M and Baum, W C. Endocrine control of prostatic carcinoma, clinical statistical survey of 1818 cases J A. M A , **143:** 1317, 1950
79 Nettleship, A and Henshaw, P S Induction of pulmonary tumors in mice with ethyl carbamate (urethane) J Nat Cancer Inst , **4:** 309, 1943
80 Ohler, R L , Houghton, J D and Moloney, W C Urethane toxicity; reports of a case of hepatic necrosis apparently due to urethane New Eng J Med , **243:** 984, 1950
81 Osgood, E E Titrated, regularly spaced radioactive phosphorus or spray roentgen of leukemias Arch Int Med , **87:** 329, 1951
82 [illegible] Leukaemia
83 [illegible] (ACTH) and cortisone in lymphomas and leukemias J A M A , **144:** 1349, 1950
84. Philips, F S Recent contributions to the pharmology of bis-(2-haloethyl) amines and sulfides Pharmacol Rev , **2:** 281, 1950
85 Philips, F S and Thiersch, J B Studies of the actions of 4 amino-pteroyl- [illegible] Exp Therap , **95:** 303, 1949
86 [illegible] tard like actions of certain [illegible] e and analogs J Pharma- [illegible]
87 Pierce, M : Lymphoma and leukemia panel Second National Cancer Conference, 1952.
88 Ragan, C , Hewes, E L , Plotz, C M , Meyer, K and Blunt, J W Effect

of cortisone on production of granulation tissue in the rabbit. Proc. Soc Exp Biol & Med , 72: 718, 1949

89 RAWSON, R W., RALL, J. E AND PEACOCK, W.: Limitations and indications in the treatment of cancer of the thyroid with radioactive iodine J. Clin Endo , 11: 1128, 1951.

90 REINHARD, E H , GOOD, J. T. AND MARTIN, E Chemotherapy of malignant neoplastic diseases. J. A M. A , 142: 383, 1950

91. RHOADS, C. P. Nitrogen mustards in the treatment of neoplastic disease J A. M A , 131: 656, 1946

92. RODGERS, C L , DONOHUE, W. L AND SNELLING, C E Leukaemia in children Canad. Med. Assoc J., 65: 548, 1951.

93 ROSENTHAL, M C., SAUNDERS, R H , SCHWARTZ, L J , ZANNOS, L., SANTIAGO, E P. AND DAMESHEK, W The use of adrenocorticotropic hormone and cortisone in the treatment of leukemia and leukosarcoma Blood, 6: 804, 1951.

94. ROSWIT, B. AND KAPLAN, G : Role of nitrogen mustard (HN_2) as a systemic adjunct to the radiation therapy of certain malignant diseases. Am J Roentgenol , 61: 626, 1949.

95. ROSWIT, B. AND KAPLAN, G : Nitrogen mustard as an adjunct to radiation in the management of bronchogenic cancer. Radiology, 57: 384, 1951.

96 RUNDLES, R W AND BARTEN, W. B : Triethylene melamine in the treatment of neoplastic disease Blood, 7: 483, 1952.

97. RUNDLES, R W., DILLON, M L. AND DILLON, E S.: Multiple myeloma. III Effect of urethane therapy on plasma cell growth, abnormal serum protein components and Bence-Jones proteinuria J. Clin Invest , 29: 1243, 1950

98 SCHULMAN, I , LANMAN, J R , LAXDAHL, D E AND HOLT, L E , JR Effects of ACTH and cortisone on leukemia in childhood Pediatrics, 8: 34, 1951.

99. SCHWARTZ, S. O. AND EHRLICH, L.: The relationship of polycythemia vera to leukemia; a critical review. Acta Hemato , 4: 129, 1950

100 SEGALOFF, A , GORDON, D , HORWITT, B N , SCHLOSSER, J. V. AND MURISON, P. J : Hormonal therapy in cancer of the breast. II. Effect of methylandrostenediol on clinical course and hormonal excretion Cancer, 5: 271, 1952.

101 SEIDLIN, S. M , ROSSMAN, I , OSHRY, E. AND SIEGEL, E.: Radioiodine therapy of metastases from carcinoma of the thyroid. a six-year progress report. J. Clin Endo , 9: 1122, 1949

102 SHIMKIN, M. B. AND BIERMAN, H R : Experimental chemotherapy of neoplastic diseases. Radiology, 53: 518, 1948.

103 SHIMKIN, M B., METTIER, S. R. AND BIERMAN, H R Myelocytic leukemia an analysis of incidence, distribution, and fatality, 1910-1948 Ann Int. Med , 35: 194, 1951.

104 SILVERBERG, G M AND DAMESHEK, W : Use of triethylene melamine in treatment of leukemia and leukosarcoma J. A. M. A , 148: 1015, 1952.

105 SKIPPER, H. E. AND BRYAN, C. E. Carbamates in the chemotherapy of leukemia. III. The relationship between chemical structure and antileukemic action of a series of urethan derivatives J Nat Cancer Inst , 9: 391, 1949.

106 SMITH, T R., JACOBSON, L O , SPURR, C L , ALLEN, J G AND BLOCK, M. H.: A coagulation defect produced by nitrogen mustard Science, 107: 474, 1948

107 SOUTHAM, C. M., CRAVER, L. F., DARGEON, H W AND BURCHENAL, J H.: A study of the natural history of acute leukemia with special reference to the duration of the disease and the occurrence of remission Cancer, 4: 39, 1951.

108 SPURR, C. L., SMITH, T. R , BLOCK, M AND JACOBSON, L. O.: A clinical study

of the use of nitrogen mustard therapy in polycythemia vera. J. Lab. & Clin. Med , 35: 252, 1950.

109 Spurr, C L , Smith, T. R., Block, M. and Jacobson, L O.: The role of nitrogen mustard therapy in the treatment of lymphomas and leukemia. Am. J. Med , 8: 710, 1950

110. Stock, C. C.. Aspects of approaches in experimental cancer chemotherapy. Am. J. Med , 8: 658, 1950

111. Stock, C. C., Biesele, J. J , Burchenal, J H , Karnofsky, D A , Moore, A. E. and Sugiura, K.: Folic acid analogs and experimental tumors Ann New York Acad. Sci., 52: 1360, 1950.

112. Stock, C C , Buckley, S , Sugiura, K and Rhoads, C. P : A comparison of the retardation of sarcoma 180 by SK 1424, 3-bis(B-chloroethyl)amino-methyl-4-methoxymethyl-5-hydroxy-6-methylpyridine with that by HN2, methyl bis-(B-chloroethyl) amine. Cancer Res , 11: 432, 1951.

[illegible]

ton Co , Phila , 1951

114. Strauss, B , Jacobson, A S , Berson, S. A , Bernstein, T C., Faden, R S and Yalow, R. S *The effect of cortisone on Hodgkin's disease* Am J Med , 12: 170, 1952.

115. Stroebel, C. F., Hall, B. E and Pease, G L : Evaluation of radiophosphorus therapy in primary polycythemia J A. M A , 146: 1301, 1951

[illegible] of 196 cases

[illegible] oprionate on the peripheral blood and bone marrow of women with advanced inoperable carcinoma of the breast Preliminary report J. Clin. Endo , 9: 666, 1949

118 [illegible]

119 Timmis, G , Galton, D and Haddow, A Personal communication

120 Treves, N Castration as a therapeutic measure in cancer of male breast. Cancer, 2: 191, 1949

121. [illegible]

122 [illegible]

123 [illegible]

124. [illegible]

Index